

Retention of antibacterial activity and bacterial colonization of antiseptic-bonded central venous catheters

A. Bach^{a,†}, H. Schmidt^a, B. Böttiger^a, B. Schreiber^b, H. Böhler^a, J. Motsch^a, E. Martin^a and H. G. Sonntag^b

^aDepartment of Anaesthesiology; ^bInstitute of Medical Microbiology, University of Heidelberg, Im Neuenheimer Feld 110, D-69120 Heidelberg, Germany

We determined how long antiseptic impregnation with silver sulphadiazine and chlorhexidine (SSC) on polyurethane central venous double- or triple-lumen catheters is retained *in vivo*. A total of 116 antiseptic catheters were tested for antibacterial activity in an *in-vitro* bioassay after various periods of *iv* catheterization. Segments from the subcutaneous (*sc*) and intravenous (*iv*) portions of the catheters were cultured. The results of test antiseptic catheters were compared with those from 117 noncoated control (*c*) catheters. Retention of antibacterial activity followed an exponential curve and lasted for up to 520 h after catheter insertion. Significant differences ($P = 0.0001$) between SSC and C catheters were noticed with regard to the quantitative level of bacterial colonization (SSC-*sc* 87 ± 34 vs C-*sc* 584 ± 122 ; SSC-*iv* 52 ± 17 vs C-*iv* 286 ± 57 ; all values are given as mean cfu \pm S.E.M.), and the frequency of bacterial colonization (SSC-*sc* 20.7% vs C-*sc* 38.5%, $P = 0.0047$; SSC-*iv* 18.1% vs C-*iv* 30.8%, $P = 0.0361$). There was no significant difference between the incidence of catheter-related bacteraemia in the test ($n = 0$) and control groups ($n = 3$) ($P = 0.2573$). Further prospective studies are required to delineate the role of antiseptic catheters in preventing catheter-related infections.

Introduction

Central venous catheters are widely used for the administration of fluids, drugs, and total parenteral nutrition, and infection related to the use of *iv* catheters continues to be a serious adverse effect, particularly in critically ill patients (Eykyn, 1984). The reported incidence of bacteraemia related to the use of central venous catheters is about 5% (Richet *et al.*, 1990). One of the measures used to control and thereby reduce the incidence of catheter-related infection is strict adherence to catheter protocols (Maki, 1989). It has recently been suggested that the use of catheters impregnated with the antiseptic agents silver sulphadiazine and chlorhexidine (SSC) reduces the risk of catheter-related infection (Maki *et al.*, 1988; Maki *et al.*, 1991; Clemence *et al.*, 1994; Ramsay, Nolte & Schwarzmann, 1994). The aim of our study was to determine the level of antimicrobial activity that was retained on the surface of these catheters after various

Tel: +49-6221-56-6355; Fax: +49-6221-56-5627

periods of iv catheterization, and to analyse the bacterial colonization of control and test catheters.

Materials and methods

Catheters

The control catheter used was the standard double- or triple-lumen polyurethane catheter manufactured by Arrow Intl., Inc. (Reading, PA). The test catheter was a similar one and was also provided by Arrow Inc., but had been treated with antiseptic bonding on the external but not luminal surface. Both types of catheter were identical in appearance; i.e. all had the same yellowish colour and not the blue coating that distinguishes the antiseptic catheter (Arrowgard, Arrow Intl., Inc., Reading, PA) that is now commercially available in the USA.

Patients and study design

After the study had been approved by the Ethics Committee of the University of Heidelberg, and informed consent obtained from the patients, a total of 57 double-lumen central venous catheters (DLC) and 59 triple-lumen central venous catheters (TLC), all of them impregnated with SSC, were inserted in 116 patients. As a control (C) a total of 117 patients received either a double-lumen (C-DLC; $n = 59$) or a triple-lumen catheter (C-TLC; $n = 58$) without any antiseptic surface treatment.

All those enrolled in the study were scheduled for cardiac surgery, and all were non-pregnant, non-lactating patients over the age of 18 years who were due to receive a DLC or TLC. Patients were excluded if there was a history of adverse reactions to silver, sulphonamides, or chlorhexidine, as were those with immune deficiency. Patients who needed additional intravascular access (with the exception of an arterial line) were also excluded.

According to the randomisation schedule, either an SSC catheter or a C catheter was inserted into the jugular vein. Catheters were inserted using the Seldinger technique after the insertion site had been cleansed with an alcoholic disinfectant with full barrier precautions (hand disinfection, sterile gloves, sterile gown, mask, hat, and large sterile drapes around the insertion site). After catheter insertion, a dry gauze dressing was applied and changed every 72 h, or earlier if soiled. Location of the catheter tip in the superior vena cava was documented by chest radiography.

A standard protocol was followed for care of the catheters. Concomitant medication used before and during the study was recorded. The decision to remove the catheter was made independently by physicians in the intensive care unit who were not involved in this study. In patients receiving antibiotic therapy, the catheter was withdrawn shortly before the next dose of the antibiotic was given.

Data collection

The medical records of all patients were screened for the following variables: age; sex; chronic underlying diseases, e.g. diabetes mellitus; allergies; type of antibiotic used for prophylaxis; use of antibiotics in the postoperative period; elective or emergency procedure; type of surgery, e.g. cardiac valve replacement or coronary bypass grafting;

duration of surgery and extracorporeal circulation; perioperative blood transfusions; duration of postoperative stay in the intensive care unit; duration of catheterization; number of intravascular devices; and adverse reactions to the test catheter, e.g. local inflammation. All data were collected on a standardised form.

Processing of catheters

After removal, the catheters were placed in a sterile container and transported immediately to the microbiological laboratory. They were then cut into 1 cm long segments under a laminar air flow hood. The tip segment and a subcutaneous (sc) segment were taken to assess bacterial colonization using a sonication technique described previously (Bach *et al.*, 1994). Segments were transferred into a tube with 5 mL brain-heart infusion (BHI) broth (Merck Diagnostics, Darmstadt, Germany) and sonicated for 3 min. One millilitre of the sonication broth and 50 μ L of serial dilutions were then placed on sheep-blood agar plates (Merck Diagnostics, Darmstadt, Germany). The number of cfu was read after aerobic incubation at 37°C for at least 24 h (see Figure 1). The limit of detection with this method was 5 cfu/catheter segment. The catheter segment was removed from the sonication broth, washed with sterile

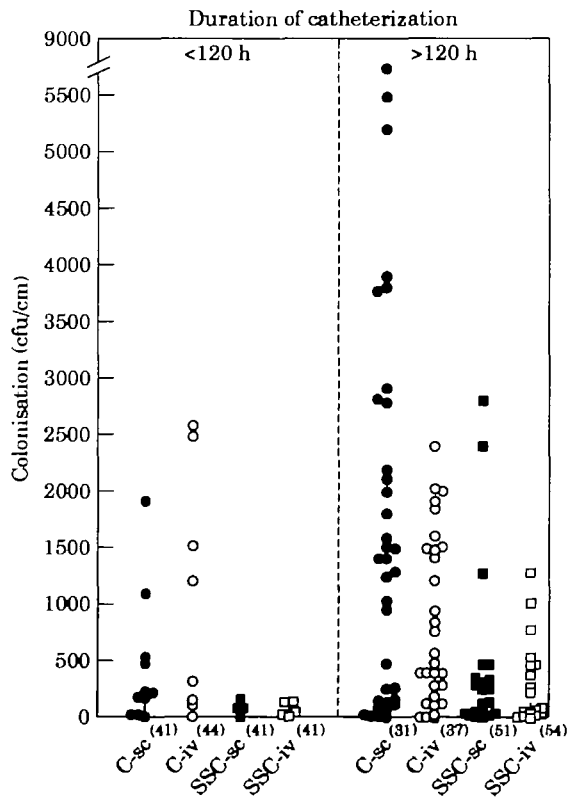


Figure 1. Bacterial colonization of catheter segments. For the sake of clarity, sterile catheters are shown as the zero mark on the vertical axis, and the number of catheter segments in brackets (*n*). Data are divided into results of catheters with durations of catheterization less and greater than 120 h. Control (C, ● ○) and test (SSC, ■ □) catheters, subcutaneous and iv segments, respectively.

phosphate buffer solution (PBS), cultured in 10 mL of BHI broth for at least 24 h and checked for growth in order to detect bacterial colonization less than 5 cfu/catheter segment. A solution of inactivating substances was added to all culture media used for quantitative culture in order to neutralize the residual activity of the antiseptic substances (Bach *et al.*, 1994). Finally, a remaining 1 cm portion of the iv part of the catheter and 1 cm of the sc segment were thoroughly washed to remove any adherent bacteria. A tube containing one of these segments and 10 mL BHI broth was capped and inverted five times, and the catheter segment was transferred to another test tube with sterile BHI broth. This procedure was repeated ten times. These catheter segments were then placed on agar plates containing antibiotic media no. 1 (Difco, Detroit, MI) inoculated with a suspension of *Staphylococcus epidermidis* ATCC 35984 to achieve a concentration of 10^6 bacteria/mL agar. Agar plates were incubated aerobically for 24 h at 37°C, and the resulting inhibition zones were measured. Uncoated catheters were used as controls.

Definitions and statistical analysis

The in-vitro antimicrobial activity of sc and iv test catheter segments against *S. epidermidis*, given as the zone of inhibition in millimetres, was correlated with the duration of iv catheterization in hours. Data were analysed using the SAS-Stat program (Release 6.03, Statistical Analysis System, SAS, Institute Inc., Cary, NC) on a microcomputer. Pairs of data are plotted (Figure 2(a), (b)) and an exponential decay function was applied.

The incidence of catheter colonization, defined as growth of ≥ 1 cfu from the 1 mL aliquot of the sonication broth and positive growth in the broth culture of the catheter segment, was compared in the test and control groups using the chi-square test with Yates' correction. The extent of bacterial colonization in the two groups, quantified by the numbers of cfu per centimetre catheter segment obtained in the cultures of the sonication broth, was compared using the Student's *t*-test (two-tailed). Bacteraemia was considered to be catheter-related if the same microorganism was isolated from culture of the catheter tip and from blood collected by venipuncture. Statistical significance was accepted at $P < 0.05$.

Results

Clinical data

For all clinical variables detailed above, no statistically significant differences were found between patients who had SSC or C catheters inserted. In particular, no allergic reaction was noticed in any patient.

Catheter colonization

The incidence and extent of catheter colonization are given in the Table and Figure 1. Only *S. epidermidis* ($n = 45$) was cultured from a total of 332 SSC catheter segments, whereas *Staphylococcus aureus* ($n = 9$), *Serratia marcescens* ($n = 6$), and *Stenotrophomonas maltophilia* ($n = 3$), in addition to *S. epidermidis* ($n = 63$) were isolated from a total of 334 C catheter segments.

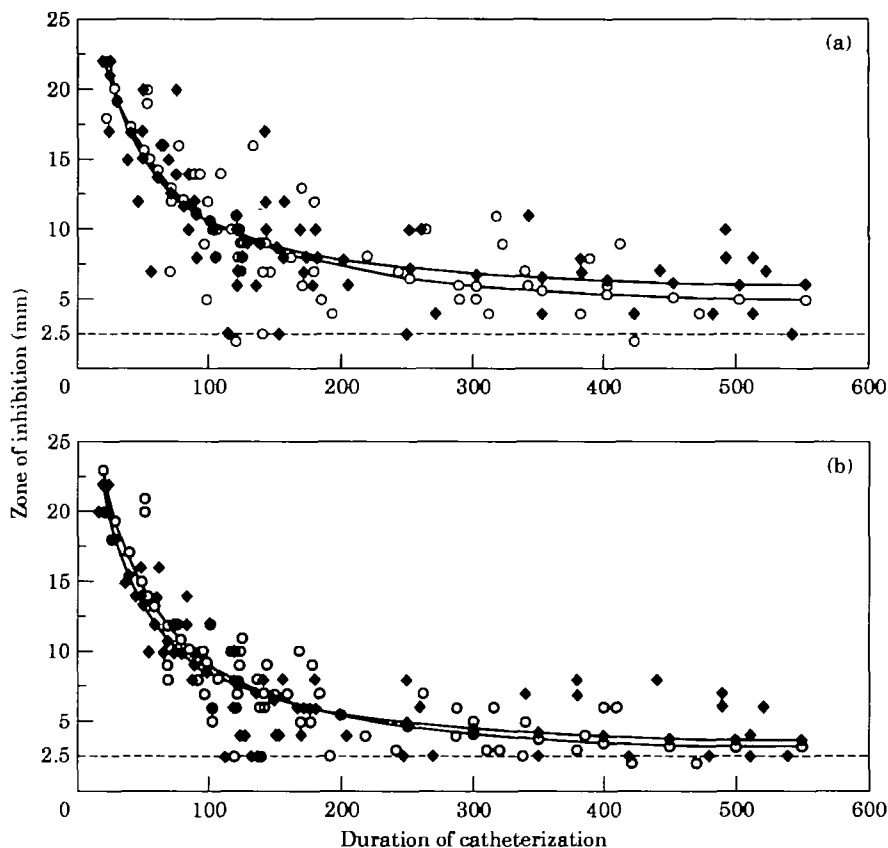


Figure 2. Zone of inhibition (mm) in relation to duration of catheterization (h) for the segments of test (SSC) double- (○, DLC) or triple-lumen (◆, TLC) catheters. The diameter of the hydrated catheter on the plate is 2.5 mm (see horizontal dashed line: this represents no inhibition of growth of *S. epidermidis*) (a) Subcutaneous segments (sc). Data pairs, $r^2 = 0.572$ for SSC-DLC and $r^2 = 0.574$ for SSC-TLC. (b) Intravenous segments (iv) Data pairs, $r^2 = 0.716$ for SSC-DLC and $r^2 = 0.753$ for SSC-TLC

Table. Bacterial colonization of catheters

| | Test (SSC) catheters | | Control (C) catheters | |
|--|------------------------|------------------------|------------------------|------------------------|
| | subcutaneous segments | iv | subcutaneous segments | iv |
| Incidence of colonization (number; percent in brackets) | 27 (20.7) ^a | 21 (18.1) ^b | 45 (38.5) ^a | 36 (30.8) ^b |
| Level of colonization (mean ± S.E.M. of cfu/cm catheter segment) | 87 ± 34 ^c | 52 ± 17 ^c | 584 ± 122 ^c | 286 ± 57 ^c |

^a $P = 0.0047$, Chi-square analysis with Yates' correction.

^b $P = 0.036$, Chi-square analysis with Yates' correction.

^c $P = 0.0001$, two-tailed Student's *t*-test.

Catheter-related bacteraemia

Catheter-related bacteraemia occurred in three patients with C catheters, but in no patients with SSC catheters. In the two patients in whom *S. epidermidis* was isolated from the catheter tip segment and in the blood culture, the similarity of these microorganisms was established by comparing the pattern of the DNA fragments after restriction digestion (Sma I, Pharmacia, Freiburg, FRG) and pulse field gel electrophoresis using a CHEF DR III chiller system (BioRad, Richmond, CA) (data not shown). In one patient in whom *S. maltophilia* was isolated from the catheter tip and from the blood culture, identity of both microorganisms was assumed since the antibiogram and the API-typing (API 20 NE, Api BioMerieux) were identical. This difference in the incidence of catheter-related bacteraemia between SSC and C groups was not statistically significant ($P = 0.26$).

Zones of inhibition

The zones of inhibition of removed catheter segments in the bioassay after various intervals of catheterization are shown in Figure 2(a), (b). The mean intervals of catheterization were as follows: SSC-DLC, 180.3 ± 113.3 ; C-DLC, 183.7 ± 120.2 ; SSC-TLC, 194.4 ± 151.2 ; C-TLC, 191.9 ± 149.2 (values are given as mean hours \pm s.d.). No antibacterial activity was seen *in vitro* with unbonded, C catheter segments. The zones of inhibition of the iv segments were smaller than the zone of the sc segment of the test catheter. This difference was significant when all iv segment zones were compared with sc segment zones of all catheters ($P < 0.0001$).

Discussion

Intravenous catheters become colonized either by intraluminal contamination, haematogenous seeding from distal sites or by the migration of microorganisms from the insertion site (Maki, 1989; Mermel *et al.*, 1991). Once microorganisms have become attached to the surface of a catheter, they may be difficult to eradicate. The biofilm mode of growth renders antibiotic treatment and host defence mechanisms less effective (Maki, 1989; Zimmerli *et al.*, 1982). Current preventive strategies include strict adherence to aseptic techniques during catheter insertion and maintenance (Maki, 1989). For example, full barrier precautions during catheter insertion have been demonstrated to significantly reduce the incidence of catheter-related infection (Mermel *et al.*, 1991). In addition, antimicrobial agents can be bonded to a subcutaneously placed barrier cuff or to the whole catheter surface (Maki *et al.*, 1988; 1991).

The efficacy of silver-impregnated catheter cuffs remains controversial. Some investigators have found these devices to be effective in reducing the incidence of catheter-related infections (Maki *et al.*, 1988; Flowers *et al.*, 1989), but others noted that there was no significant reduction in catheter-related infection or catheter-related bacteraemia with prolonged catheterization (Bonawitz, Hammell & Kirkpatrick, 1991; Norwood, Hajjar & Jenkins, 1992; Babyco, Barrocas & Webb, 1993; Groeger *et al.*, 1993). These considerations led to the development of a novel device incorporating the antiseptic substances silver sulphadiazine and chlorhexidine into the entire outer surface of a central venous polyurethane catheter. Laboratory experiments demonstrated that these catheters exerted long-lasting antibacterial activity against a wide range of

pathogens (Mermel, Stolz & Maki, 1993), and animal models demonstrated their inhibition of the migration of microorganisms from the skin to the catheter tip (Bach *et al.*, 1994).

Several groups have recently reported that this novel antiseptic catheter is effective in reducing bacterial colonization with iv catheters, and in preventing catheter-related infection or bacteraemia in various patient groups (Maki *et al.*, 1991; Clemence *et al.*, 1994; Ramsay *et al.*, 1994). For example, Maki *et al.* (1991) reported a significant reduction in the incidence of catheter-related bacteraemia (from 4.7% to 1%) when catheters bonded with SSC were used in critically ill patients. To date, however, little information has been provided on the actual retention of the antimicrobial substances on the catheter *in vivo*. Preliminary data from one clinical trial indicate that residual antibacterial activity remains after a mean of 145 h of catheter placement (Maki *et al.*, 1991). In-vitro experiments showed that the antimicrobial activity of the antiseptic catheter was minimally affected by incubation in serum for 48 h (Mermel *et al.*, 1993). Also, in an animal model, the antimicrobial activity of the antiseptic catheters was retained after catheterization for seven days (Greenfeld *et al.*, 1995).

In the present study, some iv catheter segments produced zones of inhibition in the bioassay with *S. epidermidis* after catheterization periods of up to 520 h, while others produced no zone after 120 h. A decrease in the antimicrobial activity was rapid within the first 72 h, and slow after this period. Intravenous segments of catheter produced significantly smaller zones of inhibition than sc segments, indicating the more efficient dispersal of antiseptic substances into the bloodstream. However, the antibacterial activity was retained by these iv segments for at least 120 h and by some for up to 520 h after insertion.

Our results show that SSC-bonded catheters were effective in reducing catheter related bacterial colonization, which was significantly lower and occurred less frequently in DLCs and TLCs bonded with silver sulphadiazine and chlorhexidine than in C catheters. These results are consistent with the prolonged antibacterial activity on the surface of antiseptic-bonded catheters. However, the definitive role of these devices needs to be clarified by further prospective clinical trials. We were unable to demonstrate that the difference in the incidence of catheter-related bacteraemia between the control ($n = 3$) and test groups ($n = 0$) was statistically significant ($P = 0.26$). This may, however, reflect limitations in our study, for example sample size, to detect such a difference.

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