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Electrically Conductive Catheter Inhibits Bacterial Colonization

Hayet Amalou, MD¹, Ayele H Negussie, MSc.¹, Ashish Ranjan, PhD¹, Lucy Chow, MD¹, Sheng Xu, PhD¹, Craig Kroeger³, Ziv Neeman, MD^{1,4}, Naomi P. O'Grady, MD², Bradford J Wood, MD¹

¹Center for Interventional Oncology, Clinical Center and National Cancer Institute, National Institutes of Health, Bethesda, MD

²Critical Care Medicine Department, Clinical Center, National Institutes of Health, Bethesda, MD

³VitalDyne Inc, Cokato, MN

⁴Radiological Associates, Philadelphia, PA

Abstract

Purpose—To design, prototype and assess a custom vascular access catheter for its ability to inhibit bacterial colonization *in vitro* and to optimize electrical parameters for efficacy and safe translation.

Materials and Methods—A vascular access catheter with conductive elements was designed and custom fabricated with two electrodes at the tip, separated by a non-conductive segment. The catheter was colonized with *Staphylococcus aureus* and incubated at pre-determined current levels (4-8 microAmperes (μ A) and durations (4-24hr). Catheters were compared with bacterial counts and Scanning Electron Microscopy.

Results—Bacteria colony forming units (CFU's) were reduced significantly ($p < 0.05$) by $> 90\%$ (91.7-100%) in all uninterrupted treatment arms that included electrical current (4 or 8 μ A) of at least 8 hours duration. In addition qualitative analysis using SEM revealed that the treated catheter exposed to electrical current had markedly less bacteria compared to the untreated one.

Conclusion—This catheter with conductive elements inhibits bacterial colonization *in vitro* when very small electrical current (4 to 8 μ A) is applied across the tip for 8 to 24 hours. *In vivo* validation is requisite to future translation.

Introduction

Central venous catheters (CVCs) can be associated with dangerous and costly complications, including catheter-related bloodstream infections (CRBSIs) [1]. CVCs are widely implanted and in the US alone, over 5 million catheters are in use annually [2].

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The lethal effects of electric current and electrochemical potentials to microorganisms have been documented for many decades [3-7]. However, clinical applications remain unexplored due to absence of patient compatible devices with integrated power sources, and lack of knowledge on the safe electrical parameters (duration and current) to avoid arrhythmias while maintaining bactericidal effects.

Catheter-related infections are often caused by coagulase-negative *S.aureus* and *Enterococci*, among others [8]. A major hurdle in confronting this infectious disease is the formation of protective matrix or biofilm on the CVC [9]. These biofilms promote a community of microbes embedded in an adhesive glycopolymeric matrix, and are an adapted survival mechanism to change the interaction dynamics with antibiotics and challenge host defense mechanisms [10]. They are formed on the surface of the catheter by the reversible adherence of bacterial colonies which eventually adhere irreversibly by binding to the catheter surface using exopolysaccharide glycocalyx polymers, and formation of more stable biofilms [11]. These glycocalyx polymers are produced by the microorganisms themselves, and serve as an adhesive protective “force field” for the bacteria, consequently reducing the therapeutic effect of antibiotics [10, 12]. Biofilm infections are rarely resolved by host defense mechanisms. Antibiotic therapy typically reverses the symptoms caused by planktonic cells released from the biofilm, but fails to eliminate the sequestered biofilm, isolated from usual treatments. For this reason biofilm infections typically show recurring symptoms, despite cycles of antibiotic therapy. Even in individuals with excellent cellular and humoral immune reactions, biofilm infections are rarely resolved by the host defense mechanisms. Therefore, biofilm formation may be prevented by various strategies other than the use of antibiotic therapy.

Historically, CRBSIs have been a significant public health problem as the 13th leading cause of death in the USA with an estimated total annual incidence of 250,000 cases [13, 14]. The cost for CRBSI was estimated as greater than \$56,000 per incidence [15], indicating the broad economic impact. Device strategies that could reduce the incidence of CRBSIs should be inexpensive, easy to implement, and have impact on CRBSIs across broad groups of patients.

In this study, the effects of electrical current on bacterial adherence and growth on catheter surface by applying low-amperage (4-8 μ A) direct current (DC) were investigated. Leakage of electrical current in cardiac ablation catheters should remain below 10 μ A to avoid arrhythmias. This level of current is used by electrophysiology device manufacturers as a safety threshold [16]. Furthermore, the effect of the duration of DC exposure on catheter colonization was determined.

Materials and Methods

Catheter

Custom sterile electrical conducting catheters were designed and fabricated (VitalDyne Inc, Cokato, MN) with conductive elements from the hub to internal electrodes near the distal tip. The main shaft of the catheter is insulated with Pebax ® (polyether block amide copolymer, Arkema, Cary, NC) with a negative charged 4 mm long electrode ring 45 mm

proximal to the tip, and a positive charged 4 mm electrode ring just 3 mm proximal to the distal tip, with an uninsulated conductive coating along 32 mm of the 40 mm space between the negative and positive electrodes. Electrode rings were made of platinum iridium alloy, with partially intervening conductive coating in between rings. This forces direct current to be externalized along the 8 mm non-conductive coated segment between the electrode rings. Although not specifically verified, this current should also travel along the thin layer of blood or saline contacting the surface of the catheter, since there are not conductive elements at that segment. The conductive elements receive predetermined current upon connection with a voltage generator and an ammeter that measure direct current.

Bacterial Culture and *In Vitro* Infection Model

Catheter infection was performed with live *Staphylococcus aureus*. The *S. aureus* used was a virulent capsular serotype 8 pathogen. Prior to each treatment, *S. aureus* was cultured overnight in 10 ml of Luria Bertani (LB) Broth (KD Medical Inc. Columbia MD) at 37 °C in ambient air overnight with shaking. A one-compartment *in vitro* infection model was established (Fig. 1). Briefly, 800 mL of LB broth was added into nine independent pre-sterilized canisters (Cardinal Health, Mannford, OK) at 37 °C. Following this, an ethylene oxide sterilized conducting catheter tip was inserted into each canister and shaken for a predetermined time (4, 8 or 24 hr) at 37 °C, while connected to stable electrical current at predetermined levels (Figure 1). The inserted catheters were connected to a two-channel current generator (1761 DC power supply, BK Precision, Yorba Linda, CA) supplying either no current, 4 μ A or 8 μ A direct current (DC) monitored with a Multimeter (5491A 50,000 Count True RMS Bench Digital multimeter, BK Precision, Yorba Linda, CA). Control catheters (n = 3) were treated identically, but without electrical current.

Bacterial Inoculation and Incubation- Experimental Design

After securing the conducting catheter into the canister and obtaining a stable DC reading (0, 4 or 8 μ A), the canisters were placed on temperature controlled incubator shaker (MaxQ 4000, Thermo Scientific, Waltham, MA). Subsequently, the culture was inoculated with 1 ml of standardized inoculums (1000 colony-forming unit/ml (CFU/ml)) of *S. aureus* prepared in LB broth. Current treatments were performed as shown in Table 1.

Quantitative Analysis of Treatment Effect

At the end of the treatment, each catheter from various treatment group (Table 1) was withdrawn from the canister, gently rinsed with sterile Phosphate Buffered saline (PBS) (Life Technologies, Carlsbad, CA) and then placed in a 15 ml tube containing 7 ml of PBS. The tube was placed in a water bath sonicator (5510 Branson ultrasonic cleaner, Branson Ultrasonics Corp., Danbury, CT) for a total of 5 times each for five seconds exposure time to detach adhered bacteria from the conducting catheter. Dilutions were made (up to 10^5) in PBS and samples were plated on LB plates and incubated overnight at 37 °C. CFU/ml in the tube washings were determined by manual counting after adjustment for dilution, to yield CFU per ml.

Qualitative Analysis of Treatment Effect by Scanning Electron Microscopy (SEM)

Two catheters were inoculated and incubated as previously described, and studied solely for the purpose of qualitative assessment for relative amount of adherent bacteria using scanning electron microscopy. After 24 hours incubation, the catheters were removed from the culture canister, the non-conducting catheter was cut-off and then the tip were suspended in 50 ml tubes containing 25 ml 10% (wt./v) buffered formaldehyde in PBS and kept at 4 °C until SEM imaging. The two catheters were processed for SEM according to previously established method by Farb *et al.* [17]. Low power images were acquired at 15x magnification and assembled into a montage. Regions of interest were photographed at incremental magnifications.

Statistical Analysis

Treatment groups (Table 1) were compared each to the control group for differences in mean CFU count using a t-test. All analyses were performed using GraphPad Prism 5.0 (GraphPad Software Inc.). A p-value less than 0.050 indicated statistical significance. Values are reported as mean $\pm$ SEM unless otherwise indicated. Control groups were used as reference of comparison. No three way comparisons or comparisons of the experimental groups to each other were performed.

Results

The effect of direct current on bacterial colonization of the conducting catheter was evaluated from the treatment protocols described in Table 1 and calculated colony forming units/ml (CFU/ml) counts are described (Table 1). Analysis of continuous 24 hour treatment was performed at current settings of 4 and 8 μ A. As shown in Table 1, continuous exposure of 4 and 8 μ A for 24 hours produced >90 % reduction in viable bacterial counts on the conducting catheters compared to untreated catheters with p values 0.0021 and 0.0001, respectively. Application of higher current level (8 μ A) did not result in proportional decrease in bacterial count, thereby indicating that low current level (4 μ A) was sufficient to prevent bacterial colonization *when exposed continuously for 24 hours*. Continuous exposure of 4 and 8 μ A for 8 hours also produced >90 % reduction in viable bacterial counts on the conducting catheters compared to untreated catheters with p values 0.0398 and 0.0367, respectively. These results also indicated that a continuous exposure was more effective compared to pulsed (8 hours current on and 16 hours current off) exposure of low level current. When 4 μ A currents were applied at 33% duty cycle, the reduction was less effective, but p value for this pulsed group versus controls did not reach significance (p = 0.0955).

In order to determine minimum duration of current exposures to prevent bacterial colonization, 4 hour current duration was studied. The number of bacteria attached to catheters as determined by the CFU/ml counting method was not significantly reduced (<25%, p= 0.095) in the 4 hour group compared to the control catheter. However, treatments 8 hours and longer resulted in reduction of bacterial counts compared to the untreated control (Table 1). Eight hours of continuous electrical application (4 or 8 μ A) reduced bacterial growth on the catheter, under a static *in vitro* environment compared to the 4 μ A

current for duration of 4 hours. Thus, 8 hours was defined as a threshold duration for the 4 uA current.

Discussion

Catheters impregnated with different antibiotics can reduce microbial adherence and colonization on the catheter [18, 19]. Many catheters containing antibiotics [20-22], antiseptics [23] or metals [24] have been evaluated experimentally or in clinical trials [25], and some are commercially available for use in clinics [26]. Once the biofilm is established, systemic clearance requires 2500 times higher dose of antibiotic than needed to kill non-biofilm bacteria [27]. Nevertheless, CRBSIs continue to be treated with combinations of antibiotics or more often by removing the catheter [28]. Persistent resistance is due to poor drug penetration to the full depth of the biofilm. Drug resistance may be attributed to increased cellular density [29], initiation of drug efflux pumps [30], changes in β -1,3-glucan content [31] and over expression of exopolysaccharide glycocalyx [32]. Both β -1,3-glucan and exopolysaccharide glycocalyx sequester the drug and the later imparts negative surface charge to the microorganisms [32].

Studies have shown that these negative surface charges can be disrupted by low amperage electric current ($10 \mu\text{A}$) [33]. Such disruptions can reduce bacterial colonization and also facilitate antimicrobial penetration in biofilms [12, 34]. It is speculative if the mechanism of action of this conductive catheter is related to, direct bactericidal effect, impairment of the biofilm glycocalyx or simple charge repellant to biofilm or to negatively charged bacteria themselves. Biofilm, thrombus, fibrin sheath, bacterial colonization, and CRBSI are a continuum of often-related phenomena. Regardless of the mechanism, reduction or inhibition of bacterial growth, adhesion, or colonization on catheters could be critically important for immune-compromised or intensive care patients, who may be at higher risk for infection.

In the present study, electrical current kills or inhibits adherence or growth of clinically relevant bacteria using a custom fabricated catheter with conductive elements exposed near the distal catheter. The high efficacy of preventing the bacterial adherence in this model suggests that this technology and method could theoretically inhibit bacterial colonization from skin to catheter tip or from bloodstream to catheter tip, although this is speculative. Inhibition of colonization should lead to fewer CRBSIs, and the remarkable logarithmic degree of bacterial reduction seen here is promising.

Many limitations exist in the current study related to methodology and ability to extrapolate these findings. It is unclear whether this one-compartment static model results will resemble or translate to a dynamic fluid phase *in vivo* environment. Whether short term reduction in bacterial counts will translate into meaningful clinical outcomes also remains speculative.

In general, the overall potential benefits must outweigh any associated increased costs of prophylactic technologies. Effective but expensive technology for prevention of CRBSI or colonization could be cost effective if applied only in certain high-risk settings. Although

treatment with anti-infective agents may be effective at reducing CRBSI, widespread adoption has not occurred. [35].

Low current electricity can inhibit bacterial growth on custom catheters *in vitro*. Although these early results encourage further study, potential clinical roles remain speculative. Clinically effective and cost-effective solutions to avoid CRBSI remain high impact for this common and costly problem.

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References

1. Raad I. Intravascular-catheter-related infections. *Lancet*. 1998; 351:893–898. [PubMed: 9525387]
2. Mermel LA, Farr BM, Sherertz RJ, et al. Guidelines for the management of intravascular catheter-related infections. *Clinical Infectious Diseases*. 2001; 32:1249–1272. [PubMed: 11303260]
3. Rosenber B, Vancamp L, Rosenber B, Vancamp L, Krigas T, Krigas T. Inhibition of cell division in *escherichia coli* by electrolysis products from a platinum electrode. *Nature*. 1965; 205:698–699. [PubMed: 14287410]
4. Pareilleux A, Sicard N. Lethal effects of electric current on *escherichia coli*. *Applied Microbiology*. 1970; 19:421–424. [PubMed: 4909349]
5. Spadaro JA, Berger TJ, Barranco SD, Chapin SE, Becker RO. Antibacterial effects of silver electrodes with weak direct-current. *Antimicrobial Agents and Chemotherapy*. 1974; 6:637–642. [PubMed: 15825319]
6. Thibodeau EA, Handelsman SL, Marquis RE. Inhibition and killing of oral bacteria by silver ions generated with low intensity direct-current. *Journal of Dental Research*. 1978; 57:922–926. [PubMed: 281370]
7. Bolton L, Foleno B, Means B, Petrucelli S. Direct-current bactericidal effect on intact skin. *Antimicrobial Agents and Chemotherapy*. 1980; 18:137–141. [PubMed: 7416740]
8. Edmond MB, Wallace SE, McClish DK, Pfaller MA, Jones RN, Wenzel RP. Nosocomial bloodstream infections in united states hospitals: A three-year analysis. *Clinical Infectious Diseases*. 1999; 29:239–244. [PubMed: 10476719]
9. Donlan RM, Costerton JW. Biofilms: Survival mechanisms of clinically relevant microorganisms. *Clinical Microbiology Reviews*. 2002; 15:167–192. [PubMed: 11932229]
10. Anwar H, Dasgupta MK, Costerton JW. Testing the susceptibility of bacteria in biofilms to antibacterial agents. *Antimicrobial Agents and Chemotherapy*. 1990; 34:2043–2046. [PubMed: 2073094]
11. Costerton JW, Cheng KJ, Geesey GG, et al. Bacterial biofilms in nature and disease. *Annual Review of Microbiology*. 1987; 41:435–464.
12. Costerton JW, Ellis B, Lam K, Johnson F, Khoury AE. Mechanism of electrical enhancement of efficacy of antibiotics in killing biofilm bacteria. *Antimicrobial Agents and Chemotherapy*. 1994; 38:2803–2809. [PubMed: 7695266]
13. National center for health statistics. Monthly vital statistics report. 1995; 43:5–7.
14. Pittet, D. Nosocomial bloodstream infections. In: Wenzel, RP, editor. *Prevention and control of nosocomial infections*. Baltimore: Williams and Wilkins; 1997. 711–769.
15. Dimick JB, Pelz RK, Consunji R, Swoboda SM, Hendrix CW, Lipsett PA. Increased resource use associated with catheter-related bloodstream infection in the surgical intensive care unit. *Archives of Surgery*. 2001; 136:229–234. [PubMed: 11177147]
16. Weibell F. Electrical safety in the hospital-1974. *Annals of Biomedical Engineering*. 1974; 2:126–148.

17. Farb A, Tang AL, Shroff S, Sweet W, Virmani R. Neointimal responses 3 months after p-32 beta-emitting stent placement. *International Journal of Radiation Oncology Biology Physics*. 2000; 48:889–898.
18. Raad I, Darouiche R, Dupuis J, et al. Central venous catheters coated with minocycline and rifampin for the prevention of catheter-related colonization and bloodstream infections - a randomized, double-blind trial. *Annals of Internal Medicine*. 1997; 127:267–274. [PubMed: 9265425]
19. Johnson JR, Johnston B, Kuskowski MA. In vitro comparison of nitrofurazone- and silver alloy-coated foley catheters for contact-dependent and diffusible inhibition of urinary tract infection-associated microorganisms. *Antimicrobial Agents and Chemotherapy*. 2012; 56:4969–4972. [PubMed: 22751541]
20. Hampl J, Schierholz J, Jansen B, Aschoff A. In-vitro and in-vivo efficacy of a rifampin-loaded silicone catheter for the prevention of csf shunt infections. *Acta Neurochirurgica*. 1995; 133:147–152. [PubMed: 8748757]
21. Bayston R, Fisher LE, Weber K. An antimicrobial modified silicone peritoneal catheter with activity against both gram positive and gram negative bacteria. *Biomaterials*. 2009; 30:3167–3173. [PubMed: 19289248]
22. Schierholz J, Jansen B, Jaenicke L, Pulverer G. In-vitro efficacy of an antibiotic releasing silicone ventricle catheter to prevent shunt infection. *Biomaterials*. 1994; 15:996–1000. [PubMed: 7841297]
23. Quesnel LB, Alnajjar AR, Buddhavudhikrai P. Synergism between chlorhexidine and sulfadiazine. *Journal of Applied Bacteriology*. 1978; 45:397–405. [PubMed: 103874]
24. Ostendorf T, Meinhold A, Harter C, et al. Chlorhexidine and silver-sulfadiazine coated central venous catheters in haematological patients - a double-blind, randomised, prospective, controlled trial. *Supportive Care in Cancer*. 2005; 13:993–1000. [PubMed: 15834740]
25. Hampl JA, Weitzel A, Bonk C, Kohnen W, Roesner D, Jansen B. Rifampin-impregnated silicone catheters: A potential tool for prevention and treatment of csf shunt infections. *Infection*. 2003; 31:109–111. [PubMed: 12682816]
26. Casey AL, Mermel LA, Nightingale P, Elliott TSJ. Antimicrobial central venous catheters in adults: A systematic review and meta-analysis. *Lancet Infectious Diseases*. 2008; 8:763–776. [PubMed: 19022192]
27. Nickel JC, Ruseska I, Wright JB, Costerton JW. Tobramycin resistance of pseudomonas-aeruginosa cells growing as a biofilm on urinary catheter material. *Antimicrobial Agents and Chemotherapy*. 1985; 27:619–624. [PubMed: 3923925]
28. O'Grady NP, Alexander M, Burns LA, et al. Guidelines for the prevention of intravascular catheter-related infections. *American Journal of Infection Control*. 2011; 39:S1–S34. [PubMed: 21511081]
29. Perumal P, Mekala S, Chaffin WL. Role for cell density in antifungal drug resistance in candida albicans biofilms. *Antimicrobial Agents and Chemotherapy*. 2007; 51:2454–2463. [PubMed: 17502416]
30. Ramage G, Bachmann S, Patterson TF, Wickes BL, Lopez-Ribot JL. Investigation of multidrug efflux pumps in relation to fluconazole resistance in candida albicans biofilms. *Journal of Antimicrobial Chemotherapy*. 2002; 49:973–980. [PubMed: 12039889]
31. Nett J, Lincoln L, Marchillo K, et al. Putative role of beta-1,3 glucans in candida albicans biofilm resistance. *Antimicrobial Agents and Chemotherapy*. 2007; 51:510–520. [PubMed: 17130296]
32. Costerton JW. The role of bacterial exopolysaccharides in nature and disease. *Journal of Industrial Microbiology & Biotechnology*. 1999; 22:551–563.
33. Crocker IC, Liu WK, Byrne PO, Elliott TSJ. A novel electrical method for the prevention of microbial colonization of intravascular cannulae. *Journal of Hospital Infection*. 1992; 22:7–17. [PubMed: 1358949]
34. Liu WK, Tebbes SE, Byrne PO, Elliott TS. The effects of electric current on bacteria colonising intravenous catheters. *J Infect*. 1993; 27:261–269. [PubMed: 8308318]
35. Hockenhull JC, Dwan KM, Smith GW, et al. The clinical effectiveness of central venous catheters treated with anti-infective agents in preventing catheter-related bloodstream infections: A systematic review. *Critical Care Medicine*. 2009; 37:702–712. [PubMed: 19114884]

Figure 1.

In vitro catheter colonization model. A) The sterilized catheter tip inserted into an incubating canister containing bacteria in growth medium at 37 °C. B) Electric current treatments were controlled, normalized and measured using a voltage generator power supply (below) and an ammeter / multimeter (Top). C) Conductive catheter tip with metallic ring electrodes (arrows)

Figure 2. Scanning Electron Microscopy (SEM) images of catheters incubated in *S. aureus* culture for 24 hours with and without electrical current exposure (2A and 2B have same magnification). A) Untreated control catheter exposed to zero electrical current exhibits much more numerous bacteria (arrow) on the surface of the catheter tip than test catheter. B) Exposed to 24 hours of 8 µA current. Although subjective and not specifically quantified, the catheter

treated with electrical current demonstrated markedly fewer adherent bacteria (arrow) after 24 hour incubation in *S. aureus* (Note: the bacteria have spherical shape as indicated by the white arrows).

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Table 1

Logarithmic Bacterial Reduction Following Varied Electrical Parameters and Durations

Current Exposure(μ A)	Duration (hour)	Control (Log CFU/ml)	Treatment (Log CFU/ml)	Log CFU/ml Reduction	% Reduction	P-value
4	4	5.1 $\pm$ 0.1	5.0 $\pm$ 0.1	0.1 $\pm$ 0.1	25.1	0.0950*
4	8	12.7 $\pm$ 2.6	9.5 $\pm$ 0.9	3.3 $\pm$ 2.6	99.9	0.0398**
4	8 ON/16 OFF	6.3 $\pm$ 0.6	0.8 $\pm$ 0.1	1.0 $\pm$ 0.1	85.7	0.0955*
4	24	6.3 $\pm$ 0.6	5.0 $\pm$ 0.3	1.3 $\pm$ 0.2	95.4	0.0021**
8	8	12.7 $\pm$ 2.6	9.2 $\pm$ 1.0	3.6 $\pm$ 3.0	100.0	0.0367**
8	24	5.6 $\pm$ 0.3	4.6 $\pm$ 0.2	1.0 $\pm$ 0.5	91.7	0.0001**

** Statistically significant.