Effect of different antiseptic skin agents on bacterial contamination of peripheral intravenous catheters in dogs and cats

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Abstract

This study evaluated the effect of selected antiseptic skin agents (alcohol, chlorhexidine, and their combination) on the level of microbial contamination of peripheral intravenous catheters. A total of 83 animals (73 dogs and 10 cats) in which a peripheral intravenous catheter was inserted were evaluated. Prior to insertion, one of the tested antiseptic agents was used to prepare the site of catheter insertion. For each animal, microbiological examination of two skin swabs (taken before and after application of antiseptic skin agent) and a sample from the catheter after it was removed from the vessel was performed and the duration of catheter placement in the vessel was also recorded. Regarding the proportion of positive microbiological culture result, no significant difference was found between the three antiseptic agents in the rate of bacterial contamination of the skin ($P = 0.552$) or the peripheral intravenous catheter ($P = 0.597$). The intravenous catheter contamination rate tended to increase with the duration of hospitalization, although this relationship was not significant ($P = 0.309$). Bacteria isolated from contaminated catheters matched bacteria isolated from the skin of the same animal in one case only. Our results confirm the effectiveness of individual skin antiseptic agents and suggest that contamination of peripheral intravenous catheters occurs primarily from the environment during patient's hospitalization, rather than as a consequence of insufficient aseptic skin preparation at the site of catheter insertion.

Alcohol, bacteraemia, chlorhexidine, microbiology, sepsis

Microbial contamination of peripheral intravenous catheters (PIVCs) is a relatively common complication in veterinary medicine. The reported incidence of contamination ranges from 10.4% to 39.6% (Mathews et al. 1996; Seguela and Pages 2011; Guzmán Ramos et al. 2018; Matula et al. 2023; Simpson and Zersen 2023). Skin is one of the most important routes for infection and subsequent colonization of PIVCs (Seguela and Pages 2011; Matula et al. 2023). The risk of PIVC contamination can be reduced by adequately clipping the hair around the insertion site, removing gross debris from the surface of the skin, and ensuring proper use of antiseptic skin agents. The severity and clinical consequences of microbial contamination of intravenous catheters are considerably higher in patients in intensive care units, which is why particular attention is paid to proper aseptic skin preparation of the PIVC insertion site (Jones et al. 2009; Simpson and Zersen 2023).

Bacterial contamination of PIVCs and differences in the effectiveness of various antiseptic agents for surgical site preparation have already been reported (Asimus et al. 2019). However, to our knowledge, the available literature does not provide data on the comparison of individual antiseptic skin agents and their effect on the rate of bacterial contamination of PIVCs. The aim of this study was therefore to determine the effect of selected antiseptic skin agents (alcohol, chlorhexidine, and their combination) on the level of microbial contamination of PIVCs.

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Materials and Methods

In this prospective study, 83 patients (73 dogs and 10 cats) presented to the Small Animal Clinic (SAC), Faculty of Veterinary Medicine (FVM), University of Veterinary Sciences Brno (UVS), Czech Republic, were enrolled. The animals were being treated and hospitalized at the SAC Intensive Care Unit, and had a PIVC inserted. The prerequisite for inclusion in the study was that one of the tested antiseptic skin agents was used for antiseptic preparation of the insertion site. In addition, microbiological skin swabs were obtained before and after skin preparation, and the catheter had to be removed under aseptic conditions and subsequently sent for microbiological examination. Patients in whom PIVC was prematurely removed and damaged or possibly contaminated were excluded. The PIVCs were inserted into either the cephalic vein or the lateral saphenous vein. Antiseptic preparation of the skin at the insertion site was performed according to standard procedure. Following hair clipping, the skin was prepared with one of three antiseptic regimens: 1) an alcohol-based solution (ethanol, 2-propanol, Softasept N, B. Braun Medical AG, Sempach, Switzerland), 2) a chlorhexidine spray (SkinMed spray, Cymedica spol. s r.o., Hořovice, Czech Republic), or 3) a combination of chlorhexidine soap (Lifo-Scrub, B. Braun Medical AG) and an alcohol-based solution (Softasept N, B. Braun Medical AG). Intravenous catheters with an injection port (VasoVet, B. Braun, Melsungen, Germany) with a diameter of 18G-24G (depending on patient size) were used. Each catheter was then secured to the skin with an adhesive tape, a short extension tube was connected to it, and the catheter was covered with a dressing consisting of one layer of bandage and one layer of cohesive wrap (Coban). Catheters were regularly checked at least twice a day for signs of possible phlebitis. Catheters were removed before patient discharge, in case of catheter malfunction, or when signs of phlebitis (pain, local redness, fever) developed. The dressing was changed when it became loose, soiled, or damp.

Two microbiological skin swabs and one catheter sample were collected after its removal from the vessel from each dog or cat. The first swab was taken from the skin at the PIVC insertion site after hair clipping, prior to the application of the antiseptic skin agent. The second swab was taken from the same site after the antiseptic skin agent had been applied. Swabs were collected using sterile swabs moistened with sterile saline and placed into AMIES transport medium (Oxoid, Basingstoke, United Kingdom). The sample for microbiological evaluation consisted of the distal third of the intravenous part of the catheter. Immediately after catheter removal from the vessel, the distal third was collected using sterile instruments, placed in a sterile tube and sent for microbiological examination. Care was taken to avoid contact with the animal's skin or any potentially contaminated surfaces during removal. In total, 249 samples were collected and subjected to microbiological evaluation.

Skin swabs were cultured on Columbia agar (Oxoid) supplemented with 5% defibrinated sheep blood (blood agar, BA) and MacConkey agar (Oxoid). Samples were incubated at 37 °C under aerobic conditions for 48 h. Catheter samples were aseptically removed from the collection tube, cultured on BA, and then transferred to a tube containing Brain Heart Infusion broth (Oxoid) for enrichment. Enrichment was carried out at 37 °C under aerobic conditions for 24 h. After incubation, the broth was thoroughly vortexed, 100 µl was plated onto BA, and incubated at 37 °C under aerobic conditions for 48 h. The resulting colonies were visually assessed based on morphology and subsequently identified by MALDI TOF MS (Bruker Daltonics, Germany) according to the manufacturer's instructions. Any bacterial growth, regardless of the number of colony-forming units, was considered a positive microbiological culture result. The intensity of growth was evaluated semi-quantitatively using a four-point scale.

The following data were recorded: patient signalment, type of antiseptic agent used, time of PIVC insertion, time of PIVC removal, and duration of hospitalization. Dogs and cats enrolled in the study (n = 83) were divided into three groups based on the antiseptic agent used. Based on the duration of hospitalization, patients were further divided into three groups: ≤ 24 h, > 24–≤ 48 h, and > 48 h. The data obtained were statistically evaluated using Fisher's exact test and the results were considered significant at a P value of 0.05.

Results

A total of 83 animals were enrolled in the study, including 73 dogs and 10 cats, from which 249 samples were collected. The skin at the PIVC insertion site was treated with an alcohol solution in 32 animals (28 dogs and 4 cats), with chlorhexidine spray in 25 animals (23 dogs and 2 cats), and with a combination of chlorhexidine soap and alcohol solution in 26 animals (23 dogs and 3 cats). The lowest rate of skin contamination after antiseptic treatment was observed with chlorhexidine spray (1 out of 25 samples, 4.0%). The proportion of positive culture samples from individual swabs is shown in Table 1. In cases where the skin swab collected after antiseptic treatment was positive, a single bacterial species was cultured in each animal, while in one animal, multiple bacterial species were detected. The distribution of bacterial species identified in the skin swabs collected after antiseptic treatment is presented in Table 2. No significant difference was found between the three antiseptic agents regarding the proportion of positive skin swabs (P = 0.552; Table 1).

Out of the 83 PIVCs tested, 18 samples (21.7%) yielded positive cultures. In the group treated with alcohol-based solution, 7 animals (21.9%) had positive cultures; in the group treated with chlorhexidine spray, 7 animals (28.0%) had positive cultures; and in the group treated with a combination of chlorhexidine soap and alcohol-based solution, 4 animals (15.4%) had positive cultures (Table 1). When positive, catheter samples contained one or more bacterial colonies. The most commonly isolated bacterium was *Enterobacter* spp. (7 occurrences, 38.9% of all positive samples). Other frequently isolated bacteria included *Staphylococcus*, *Micrococcus*, *Corynebacterium*, *Pseudomonas*, and *Bacillus* spp. (Table 3). Bacteria isolated from contaminated catheters corresponded with those isolated from

Table 1. Proportion of positive samples identified from the microbiological examination of skin swabs collected before and after skin preparation at the peripheral intravenous catheter insertion site using one of the selected antiseptic agents (alcohol, chlorhexidine, combination of chlorhexidine soap and alcohol-based solution), and from peripheral intravenous catheters, with statistical data evaluation.

Positive sample	Alcohol	Chlorhexidine	Chlorhexidine + alcohol	Fisher's exact test
Swab before skin preparation	68.8% (22/32)	76.0% (19/25)	73.1% (19/26)	$P = 0.866$
Swab after skin preparation	12.5% (4/32)	4.0% (1/25)	7.7% (2/26)	$P = 0.552$
Catheter culture	21.9% (7/32)	28.0% (7/25)	15.4% (4/26)	$P = 0.597$
Catheter culture (≤ 24 h)	9.1% (1/11)	26.7% (4/15)	0.0% (0/9)	$P = 0.232$
Catheter culture (> 24–≤ 48 h)	40.0% (4/10)	16.7% (1/6)	11.1% (1/9)	$P = 0.426$
Catheter culture (> 48 h)	18.2% (2/11)	50.0% (2/4)	37.5% (3/8)	$P = 0.585$

Table 2. Number of bacterial isolates recovered from skin swabs collected after antiseptic skin preparation at the peripheral intravenous catheter insertion site, grouped by the type of antiseptic agent used (alcohol, chlorhexidine, or combination of chlorhexidine soap and alcohol-based solution).

Bacterial genus	Alcohol	Chlorhexidine	Chlorhexidine + alcohol	Total
Staphylococcus spp.	2	0	1	3
Bacillus spp.	2	0	0	2
Micrococcus spp.	0	1	0	1
Enterobacter spp.	0	0	1	1

Table 3. Number of bacterial isolates recovered from the distal third of peripheral intravenous catheters after removal, grouped by the type of antiseptic agent used for skin preparation prior to catheter insertion (alcohol, chlorhexidine, or combination of chlorhexidine soap and alcohol-based solution).

Bacterial genus	Alcohol	Chlorhexidine	Chlorhexidine + alcohol	Total
Staphylococcus spp.	2	3	0	5
Bacillus spp.	1	1	0	2
Micrococcus spp.	0	1	1	2
Enterobacter spp.	4	1	2	7
Leclercia spp.	1	0	0	1
Pseudomonas spp.	1	1	0	2
Corynebacterium spp.	0	2	0	2
Streptomyces spp.	0	1	0	1
Moraxella spp.	0	1	0	1
Kocuria spp.	0	1	0	1
Arthrobacter spp.	0	1	0	1
Enterococcus spp.	0	0	1	1

the animal's skin in only one case (5.6%), specifically, *Micrococcus luteus*. No significant difference in the frequency of positive catheter cultures was observed between the groups based on the type of antiseptic agent used for skin preparation ($P = 0.597$; Table 1).

A total of 35 patients (31 dogs and 4 cats) were hospitalized for ≤ 24 h. A positive microbiological result from the examination of PIVC was found in only 5 patients (14.3%, all dogs), and no significant difference in the proportion of positive samples was observed between groups based on the antiseptic agent used for skin preparation ($P = 0.232$; Table 1). In 25 patients (22 dogs and 3 cats), the PIVC was in place for > 24–≤ 48 h, and a positive microbiological result was confirmed in 6 catheters (24.0%, 5 dogs and 1 cat). Again, the proportion of positive samples did not differ significantly between the groups based on the antiseptic agent used for skin preparation ($P = 0.426$; Table 1). In 23 patients (20 dogs and 3 cats), the peripheral intravenous catheter was in place for > 48 h, with 7 positive microbiological results (30.4%, all dogs). Similarly, no significant difference in positive culture rates was observed between the groups based on the antiseptic agent used ($P = 0.585$; Table 1).

As no significant differences were found in the proportion of positive culture samples between selected groups, the effect of hospitalization duration on bacterial contamination of PIVC was assessed for all patients collectively, regardless of the antiseptic agent used. The proportion of positive samples increased with the duration of hospitalization (i.e., with the time the PIVC was in place), although this difference was not significant ($P = 0.309$; Table 4).

Table 4. Proportion of positive culture samples from peripheral intravenous catheters, categorized by the duration of patient hospitalization.

	Duration of hospitalization			
	≤ 24 h	> 24–≤ 48 h	> 48 h	Fisher's exact test
Positive catheter culture	14.3% (5/35)	24.0% (6/25)	30.4% (7/23)	$P = 0.309$

Discussion

Microbial contamination of PIVCs in patients is a significant issue in veterinary medicine (Marsh-Ng et al. 2007; Seguela and Pages 2011; Guzmán Ramos et al. 2018; Matula et al. 2023; Simpson and Zersen 2023). While a positive culture of PIVC does not necessarily lead to local or systemic infection in most patients, it can result in clinical complications in some cases, particularly in immunocompromised individuals (Maki et al. 1977; Marsh-Ng et al. 2007; Seguela and Pages 2011). Some studies have identified potential risk factors for microbial colonization of PIVCs, including intravenous administration of glucose solutions, duration of catheter placement, local complications, and administration of immunosuppressive drugs or presence of immunosuppressive diseases (Seguela and Pages 2011; Guzmán Ramos et al. 2018). However, other studies have not confirmed these associations (Mathews et al. 1996; Marsh-Ng et al. 2007; Jones et al. 2009; Matula et al. 2023), highlighting the complexity of PIVC contamination which is influenced by multiple variables. It is therefore essential not to underestimate the principles of asepsis and antisepsis to minimize the risk of microbial colonization of PIVCs (Irwin et al. 1973; Zhang et al. 2016). The aim of this study was to determine whether the choice of antiseptic agent for skin preparation influences the risk of PIVC contamination and to assess the effect of hospitalization duration on this risk. In our study, the lowest relative frequency of PIVC contamination after antiseptic treatment was observed with chlorhexidine spray, with positive cultures found in 4.0% of skin swabs. However, since no significant differences were found between the three antiseptic agents in terms of positive swabs, it cannot be conclusively stated that chlorhexidine spray

significantly reduces the risk of PIVC colonization compared to alcohol-based solution or combination of both agents. Based on our findings and previous reports (Irwin et al. 1973; Guzmán Ramos et al. 2018), adherence to proper aseptic technique appears to be more important than the specific antiseptic agent used. In small animal veterinary medicine, the reported incidence of microbial colonization of PIVCs in hospitalized patients ranges from 10.4% to 15.4% (Mathews et al. 1996; Seguela and Pages 2011; Matula et al. 2023). In patients presenting with clinical signs of extravasation, catheter obstruction, or phlebitis, the incidence can increase to as much as 39.6% (Guzmán Ramos et al. 2018). The results of the relative frequency are therefore within the average values reported in other studies (Mathews et al. 1996; Seguela and Pages 2011; Guzmán Ramos et al. 2018; Matula et al. 2023).

An interesting finding of this study is that the bacteria species isolated from contaminated PIVCs corresponded with those isolated from the animal's skin in only one case, i.e., in 5.56%. According to studies in human medicine, the patient's skin is often a source of PIVC contamination (Irwin et al. 1973; Zhang et al. 2016). However, in the study by Irwin et al. (1973), in which skin swabs were collected from the site of intravenous catheter insertion both before catheter placement and after its removal, no correlation was found between microbiological findings from skin cultures and those from intravenous catheters. This was observed in patients whose skin had been antiseptically prepared with alcohol solution and a tincture of benzalkonium and solution of benzalkonium, followed by gentamicin cream application, as well as in patients in whom the intravenous catheter was inserted using a sterile technique (with sterile gloves), with or without subsequent application of gentamicin cream (Irwin et al. 1973). Thus, the use of gloves may prevent catheter contamination by the person inserting it, while emphasizing the importance of adhering to aseptic principles during catheter placement. Studies in veterinary medicine evaluating the contamination rate of PIVCs do not directly address the causal route of infection and often automatically assume the patient's skin to be the source of contamination (Seguela and Pages 2011; Matula et al. 2023). In contrast to our prospective study, these studies did not investigate the correlation between microorganisms found on the patient's skin and those detected on the intravenous catheter. Our study results demonstrated that, when proper antiseptic skin preparation is performed prior to PIVC insertion, this route of catheter contamination may be negligible. In cases where a positive microbiological result was obtained from the PIVC, the source of the infection was likely not the patient's skin, indicating it was not a consequence of inadequate antiseptic skin preparation at the site of PIVC. Other potential sources of PIVC contamination include transfer via the catheter hub, colonization by bacteria circulating in the bloodstream, or contaminated infusion solutions (Zhang et al. 2016). Our findings suggest that, in small animal medicine, these three routes represent a more significant source of PIVC contamination than the patient's skin, indicating it was not a consequence of inadequate antiseptic preparation at the site of PIVC insertion. In veterinary patients, unlike in humans, an important factor is that the animal may not cooperate with the attending veterinarian and may more frequently attempt to remove the PIVC, during which it can easily become contaminated. Therefore, to prevent PIVC infections, it is essential to focus on proper care of PIVC throughout the patient's hospitalization at the veterinary clinic.

The effect of patient hospitalization duration on the frequency of PIVC contamination has been the subject of many studies, the results of which are often conflicting (Mathews et al. 1996; Marsh-Ng et al. 2007; Jones et al. 2009; Seguela and Pages 2011; Matula et al. 2023). Previously, it was recommended that a catheter should not remain inserted in the vein for more than 48 to 72 h (Spencer 1982; Murtaugh and Mason 1989). More recently, however, it has been suggested that the timing of catheter removal should be guided by clinical indicators rather than by a fixed duration (Mathews et al. 1996;

Marsh-Ng et al. 2007). Nevertheless, the incidence of intravenous catheter contamination in patients with local clinical signs (extravasation, catheter obstruction, phlebitis) has been shown to increase with longer hospitalization, although these findings have not reached statistical significance (Guzmán Ramos et al. 2018).

The aim of our study was to verify the validity of this hypothesis and also to compare the risk of PIVC contamination over time depending on the type of antiseptic agent used for aseptic skin preparation prior to catheter insertion. However, the groups of positive catheter culture samples, stratified by antiseptic agent, did not differ significantly from each other in terms of contamination frequency. Thus, we did not demonstrate that any of the tested antiseptic agents significantly reduced the incidence of PIVC contamination during patient's hospitalization, regardless of its length. The number of culture-positive PIVC samples in our patients did show a tendency to increase with longer hospitalization (i.e. longer catheter dwell time), but this trend was not significant. In agreement with other studies (Mathews et al. 1996; Jones et al. 2009; Guzmán Ramos et al. 2018; Matula et al. 2023), we therefore confirmed that the duration of patient's hospitalization does not significantly affect the frequency of PIVC contamination in small animals.

The results of this study show that maintaining proper aseptic techniques during PIVC insertion in small animals reduces the risk of bacterial contamination of the catheter from the patient's skin to a negligible minimum, regardless of which of the tested antiseptic agent is used for skin preparation prior to PIVC insertion. Based on these findings, it can be concluded that, after *lege artis* aseptic insertion of a PIVC into the cephalic or lateral saphenous vein, the patient's skin should not be considered the primary source of bacterial contamination of PIVC in small animals. Future studies should therefore focus on identifying the main risk factors contributing to PIVC contamination. Although the number of culture-positive PIVC samples in our patients showed a trend of increasing contamination with longer hospitalization duration, this association was not significant. Thus, the PIVC dwell time should not be limited by a certain time interval but rather guided by patient's clinical signs and overall health status.

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