

Title: Elimination of *Candida albicans*' Biofilms on Intravascular Catheters  
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## Research Strategy

This paper is the product of one semester's work and research. I began the research with preliminary consultations with Professor Birthe Kjellerup and Goucher Library's reference librarians to help my investigation of microbiology, bacteriology, and public health. After searching keyword terms such as "bacterial infections", "disease", and "microbiology" and cross-referencing them with the WorldCat and the Goucher College Classic Library Catalogue, I discovered there is a great amount of recent research on *Candida*-based infections. As I skimmed through the papers, I noticed *Candida albicans* infections in catheters are significant contributors to nosocomial infections. This reinforced for me the value of not only reviewing the literature on this topic, but also the necessity of proposing an optimal strategy to reduce catheter-related bloodstream infections associated with *Candida albicans*.

I then turned to PubMed electronic database and searched for articles on the topic of eliminating bloodstream infections caused by *Candida albicans* in catheters. The reference lists of selected studies were examined for relevant articles, and efforts were made to find recent articles. Additionally, I made sure that the articles were primary literature and peer-reviewed. During this process, the research librarians assisted me in finding free full articles and using more common synonym search words to increase the number of eligible articles. Additionally, they aided me in using the "RefWorks" in order to keep a record of my references and citation.

After finishing the research, I learned that writing a quality research paper requires a lot of work and that it is helpful to start broad and then narrow down the topic. This way, the chances of finding the topic of interest and its relevance to other disciplines increases. For me, I enjoyed collaborating with my professor and the research librarians and using various library resources and databases to prepare this work. I am greatly indebted to my professor and the library resources for this review paper, which provides current success and failure of the new preventative approaches, and their comparison to the older strategies in order to understand which preventative treatment is the most effective in controlling the catheter-related BSIs.

## **Abstract**

Intravascular catheters are among the most commonly inserted medical devices which are known to cause a great number of catheter related bloodstream infections (BSIs). Biofilms are associated with many chronic infections originating from aggregation of microorganisms. One of these organisms, *Candida albicans*, has shown to be one of the leading organisms responsible for catheter-related BSIs. The presence of biofilm on intravascular catheters provide tolerance against antimicrobial treatments, thus alternative treatment strategies are sought. Originally, many strategies such as application of combined antimicrobials, addition of antifungals, and removal of catheters have been practiced that were not successful in eradicating BSIs. Since these infections result in significant morbidity, mortality, and healthcare cost, other preventive strategies including antimicrobial lock therapy, chelating agents, alcohol, and biofilm disruptors have been used that seem promising. In this review, current success and failure of the new approaches, and their comparison to the older strategies are discussed in order to understand which preventative treatment is the most effective in controlling the catheter-related BSIs.

## **Introduction**

Microorganisms mainly exist in biofilms in natural environments, which are defined as communities of microorganisms that are set in a self-produced extracellular polymeric substance (EPS) attached to a surface (Raad *et al.*, 1996; Martinez *et al.*, 2010). Organisms prefer the biofilm mode compared to the planktonic mode as they are able to exchange nutrients and genetic materials as well as provide protection to one another (Martinez *et al.*, 2010). It has become particularly evident to the medical community that biofilms are the causative agents of

nosocomial and chronic infections, many of which are resistant to current antimicrobial therapies. One of the many nosocomial infections developed in a hospital is catheter-related BSIs (Zhang *et al.*, 2013). In U.S., an approximate of 500,000 central-line associated BSIs is reported every year, however, the estimated number of infections is probably higher than reported (Maki, 2010). Due to longer hospital stay, the estimated cost of catheter-related BSIs is approximately \$33,000-\$65,000 per case (Maki, 2010). Collectively, these infections lead to high morbidity, mortality, and costs for health care delivery system.

There are a number of microbial species causing nosocomial bloodstream infections. The most common of these are *Staphylococci*, *Staphylococcus aureus*, and *Candida* species that are related to catheter-related BSIs (Lewis *et al.*, 2002). Specifically, *Candida* species are reported to be responsible for 38% rate of mortality and morbidity in BSIs (Lewis *et al.*, 2002). Among the *Candida* species, the fungus-*Candida albicans* has shown to be commonly associated with BSIs, where it forms biofilms on the surface of intravascular catheters (Martinez *et al.*, 2010). *C. albicans* are also known to cause candidiasis displaying mucosal and systemic infections (Boon *et al.*, 2008). An infection develops when microorganisms from patient's skin at the insertion site of the catheter attach to the surface of the indwelling catheter to form a biofilm (Raad *et al.*, 1996). Following the biofilm formation, the microbial cells from biofilm disperse into the bloodstream leading to serious infections (Raad *et al.*, 1996). Detachment of aggregates of these cells, their production of endotoxins, or other pyrogenic substances leads to the symptoms of disease in patients (Zhang *et al.*, 2013).

Although *C. albicans* is among the top four leading causes of catheter related BSIs, other *Candida* species such as *Candida parapsilosis*, *Candida Pseudotropicalis*, and *Candida glabrata* are capable of forming biofilm, however, they are less pathogenic compared to *C. albicans*

(Douglas, 2003). *Candida* are characterized as commensal organisms, however they can be pathogenic at times that the host defense is not fully active (Kojic & Darouiche, 2004). As an opportunistic pathogen, *C. albicans* (Lewis *et al.*, 2002) easily adapts to its surrounding environment with the help of its recognition proteins (adhesions), morphogenesis (conversion from yeast to hyphal form), and its proteolytic and lipolytic enzymes (Lewis *et al.*, 2002). They primarily infect patients who have immunocompromised state, diabetes mellitus, inserted medical device, and intravenous drug fluid feeding their body (Kojic & Darouiche, 2004). Similar to most microorganisms, *C. albicans* exist in biofilm form that not only provides a protected environment, but it also allows for horizontal gene transfer that potentially code for antibiotic resistance up to 1,000 fold greater than their planktonic counterparts (Douglas, 2003; Andes *et al.*, 2004; Zhang *et al.*, 2013). Thus, *C. albicans* have a higher resistance to antifungal and antimicrobial approaches making it difficult to prevent them from causing BSIs. Therefore, to improve patient outcome and to reduce healthcare costs, there is considerable interest in lowering the incidence of these infections and seeking potential solutions. Although this is challenging, various prevention strategies have demonstrated success that requires further research. The goal of this paper is to evaluate present prevention strategies, their limitations, and introduction of new technologies.

### **Biofilm formation and characteristics**

Development of catheter-related BSIs is associated with the biofilm formation on the device. *C. albicans* biofilms are complex and diverse microorganism communities that possess unique characteristics which need to be considered when presenting biofilm prevention solutions. *C. albicans* biofilm forms in three different stages starting when 1) these organisms attach to the surface of the catheter, 2) followed by secretion of extracellular polymers, and 3) continued by

formation of a 3-D structure that surrounds to protect the organisms (Chandra *et al.*, 2001; Kojic & Darouiche, 2004). Sometimes, contamination of the catheter surface at the time of insertion introduces organisms into the catheter lumen leading to infections (Douglas, 2003). As these biofilms mature, various morphologies and components such as polysaccharides and proteins are observed (Kojic & Darouiche, 2004). Carbohydrates are one of the main component of *C. albicans* biofilm and Chandra *et al.* (2001), confirmed in a study that addition of saliva and glucose enhanced the biofilm formation on a denture acrylic model.

Andes *et al.* (2004), described *C. albicans* biofilm in a rat model as a bilayer structure verified both by fluorescent and scanning electron microscopy which could explain the high pathogenicity caused by this organism (Douglas, 2003; Andes *et al.*, 2004; Kojic & Darouiche, 2004). Further, the inner portion of the biofilm was thin, whereas the outer portion was dense and contained both yeast and hyphal form of *C. albicans* (Andes *et al.*, 2004). In other studies, confocal laser scanning electron microscopy (CLSM) revealed *C. albicans* biofilms as a heterogeneous 3-D structure that contained water channels similar to most other biofilms (Chandra *et al.*, 2001; Douglas, 2003). While the basic characteristics of *C. albicans* biofilm in laboratory settings are similar to other species biofilm, evaluation of infected tissues exhibited multiple morphological forms of *C. albicans* including but not limited to hyphae or pseudohyphae and oval budding yeast (Douglas, 2003). These morphological aspects of the biofilm contribute to the stability of the fungal communities. Douglas (2003) reported when hypha-negative mutant was grown, it contained basal yeast layer while the yeast-negative mutant created hyphal-like biofilm similar to the wild-type ones. Later, it was observed that the yeast-negative mutants were less structurally stable indicating that the basal yeast layer assists in anchoring and supporting the biofilm onto the surface (Douglas, 2003). These biofilm

characteristics could be crucial in proposing prevention solutions to infections caused by *C. albicans* that should be characterized at their infected tissue levels.

### **Current preventative approaches and their effectiveness**

Traditionally, various antimicrobials and antifungals have been extensively used for prevention of bloodstream catheter infections. However, *C. albicans* biofilm structure and up-regulation of the resistance gene expressions allow them to be resistant to antimicrobial regimens as high as 30 to 2,000 times as that of planktonic cells (Chandra *et al.*, 2001; Lewis *et al.*, 2002; Douglas, 2003; Kojic & Darouiche, 2004). Since *C. albicans* is a commensal fungal organism, the inhibition of its fungal activity could lead to decreased infections in the bloodstream (Martinez *et al.*, 2010). Antifungal drugs have also been broadly used for the purpose of finding solution to catheter related BSIs.

The biofilm structure contributes to the exhibited tolerance of *C. albicans* to a wide spectrum of antimicrobial and antifungal agents. It is suggested that the matrix of the biofilm acts as a barrier to prevent the agents to penetrate and exert their effect on the microorganism communities (Douglas, 2003). To test this, biofilm communities that were grown in shaking conditions observed to have disrupted matrix that decreased their susceptibility (20%) to amphotericin B as opposed to those grown in static conditions (Baillie *et al.*, 1998). As mentioned earlier, the presence of a thick EPS layer as the biofilm ages contribute to the resistance level against antifungals as was seen by Kuhn *et al.* (2002) when fluconazole, nystatin, chlorohexidine, terbinafine, and amphotericin B were tested. Additionally, expression of genes that code for multidrug efflux pump could give rise to a multidrug resistant phenotype (Douglas, 2003). The CDR and MD genes encode for the two efflux pumps of ATP-binding cassette (ABC)

and other facilitators respectively (Douglas, 2003). These two efflux pumps were shown to be activated during biofilm formation and a mutation in one or both of these genes resulted in *C. albicans* susceptibility to fluconazole only in its planktonic form (Ramage *et al.*, 2002). Thus, there are multiple factors that play a role in determining whether *C. albicans* show resistance to antimicrobial and antifungal agents.

### Antimicrobials

In 1995, Raad *et al.* reported catheters that were coated with antimicrobials of minocycline and rifampin inhibited the activity of *C. albicans* in vitro compared to the catheters coated with chlorohexidine gluconate and silver sulfadiazine. The inhibitory activity was evaluated using Kirby-bauer method in which the zone of inhibition of each catheter treated with a combination of anti-infective agent was measured in mm. They described the higher efficacy of minocycline and rifampin could be due to their synergistic effect that added to their inhibition activity against *C. albicans* related infections. Additionally, minocycline-rifampin possesses a broad spectrum activity against *Candida* and a number of other bacterial species which could be useful in treating polymicrobial biofilms (Raad *et al.*, 1995). This combination also showed to have a longer half-life of antimicrobial activity of 25 days as opposed to 3 days half-life for catheters that contained chlorohexidine gluconate and silver sulfadiazine (Raad *et al.*, 1995). Even though the minocycline-rifampin combination acted as inhibitory agents against *C. albicans* biofilm, a comparative evaluation of their activity against another strategy such as chelating agent was not done. Therefore, an accurate conclusion of antimicrobial effectiveness cannot be drawn. In another study, Hanna *et al.* (2006), demonstrated that central venous catheters coated with gendine were more effective in preventing biofilm formation of *C. albicans* than those treated with antibiotics, or platinum, silver and carbon. Gendine, an antiseptic that



contains gentian violet and chlorhexidine was more effective against *Candida* species. They further mentioned that antimicrobial therapies of minocycline-rifampin have limitations when gram-negative or *Candida* species are present in catheters (Hanna *et al.*, 2006).

Observing some of the limitations of antimicrobials, more recently, Maki (2010) found that placing antimicrobial luer-activated connector coated with nanoparticle silver into the surface of catheters killed a significant amount of *C. albicans* as well as five other examined bacterial species. According to Maki (2010), application of nanoparticle silver into the catheters reduced 99.9% of *C. albicans*. Knowing that high silver concentration could negatively affect the health of individuals, one might feel cautious of this strategy. Though, Maki (2010), mentioned that the bactericidal ionic silver particles released from the surface of catheters is approximately 0.05 ng/mL that is greatly below the mean blood silver level of 0.2-5 ng/mL in an untreated healthy individual. So the silver particles in the fluid path are thought to add to the inhibitory activity of antimicrobial agents to prevent biofilm formation. Although antimicrobial agents have been used to treat intravascular infections, the challenge is that they only exhibit short term suppression and they lead to possible drug-resistance that needs to be addressed (Hanna *et al.*, 2006).

### Antifungals

In a similar arena of treating catheter-related BSIs caused by the commensal fungal organism, *C. albicans*, work has been done to evaluate the effectiveness of antifungals as a prevention solution. Later, Martinez et al. (2010) described the polymer known as chitosan, which was isolated from crustacean exoskeleton reduced the cell viability and metabolic activity in *C. albicans* biofilms in vitro, and biofilm inhibition in vivo model. As a hydrophilic polymer,

chitosan is created as a result of N-deacetylation of crustacean chitin that is able to penetrate fungal cells by damaging negatively charged cell membrane of species (Martinez *et al.*, 2010). Using caspofungin, Lazzell *et al.* (2009) was able to observe a significant reduction of the *C. albicans* amount in the catheters in vivo up to approximately  $4\log_{10}$  (Lazzell *et al.*, 2009). Among all the antifungals that have been tested, caspofungin is the most effective agent that inhibits the synthesis of *Candida* cell wall component,  $\beta$  1,3-glucan, that was observed to be damaged under confocal microscopy (Kuhn *et al.*, 2002; Douglas, 2003).

There are a limited number of antimicrobial and antifungal therapies that showed a decrease in BSIs. It was found that even though antimicrobials and antifungals seem to prevent biofilm formation in *C. albicans*, the reduction level is low and often times a regrowth and resistance is witnessed. Thus, several criteria regarding biofilm nature should be considered in order to reach an effective treatment. Since biofilms are complex and heterogenous structures, it is preferred to apply inhibitory agents that possess a wide spectrum of activity against diverse bacterial and fungal species. To avoid drug resistance, a combination of antimicrobials is suggested to use (Hanna *et al.*, 2006). Considering the ineffectiveness of these strategies, alternative treatments are sought.

### **Potential approaches**

Lack of success in treating catheter-related BSIs with antimicrobials and antifungals has urged the scientific community to propose alternative strategies. These strategies have been reported to control biofilms. Currently antimicrobial lock technique (ALT), ethanol application, chelating agents addition, and biofilm dispersion have been studied. Of these four approaches, ALT and ethanol addition can be considered the most researched ones, while more work is

encouraged to expand our knowledge on chelating agent and biofilm dispersion mechanisms and their role in inhibiting *C. albicans* biofilms in catheters.

Highly researched mechanisms: antimicrobial lock technique and ethanol application

ALT has been used to eliminate the biofilm communities consisting of gram-positive, gram-negative, and fungal cells (Toulet *et al.*, 2012). Through this technique, a high concentration of an antimicrobial solution is instilled inside the infected catheter to sterilize the catheter. Toulet *et al.* (2012) applied a concentration of 1000 mg/L of liposomal amphotericin B (L-AMB) that was able to inhibit biofilm activity of a number of *Candida* species for up to 48 hours after the end of the lock therapy. However, a complete removal of the biofilm was not obtained in this study. Although the technique mentions antimicrobial lock, other solutions such as ethanol that have inhibitory effects have also been tested. A promising study which evaluated ethanol-based and trisodium citrate (TSC) catheter lock solution against several microorganisms concluded that 60% ethanol therapy completely eradicated biofilms formed with *C. albicans* and gram-negative bacilli at 20 minutes (Balestrino *et al.*, 2009). In contrast, the 46.7% TSC only showed a decrease in *C. albicans* biofilm after 24 hours (Balestrino *et al.*, 2009). This ethanol lock solution was successful in biofilm eradication as ethanol is able to denature proteins and cause membrane leaks combined with the instilled flow of the solution.

Despite the high effectiveness of the ALT, there are a number of concerns with the approach. Sufficient care is needed when setting the antimicrobial lock method since a leak in the catheter can flush toxic concentrations of the antibiotic agent in the systemic circulation of the patients leading to toxicity and possibly death (Balestrino *et al.*, 2009). Another concern is that flushing high concentrations of strong antimicrobial agents could lead to antimicrobial

resistance (Balestrino *et al.*, 2009). Considering these concerns, ethanol could be the most optimal lock solution because of its antimicrobial activity against a broad spectrum of microorganisms, its low cost, and its absence report of resistance (Balestrino *et al.*, 2009).

Knowing the inhibitory activity of ethanol, Mukherjee *et al.* (2006) investigate the ethanol-dependent pathway in *Candida* biofilms. They reported that alcohol dehydrogenase (ADH) was down-regulated in *Candida* biofilms analyzed by proteomics, Western and Northern blotting. Further, they observed when ADH was down-regulated using disulfiram and 4-methylpyrazole, a denser *C. albicans* biofilms was formed that strengthened its ability to invade the host tissues. In case of an *adh1* mutant stain, less ethanol but more acetaldehyde was formed compared to the wild-type stains. Additionally, Mukherjee *et al.* (2006) reported that ethanol treatment was effective in reducing biofilm's biomass made by *C. albicans* ( $P < 0.05$ ), but not by *Staphylococcus spp.* ( $P > 0.05$ ) in a rabbit model of catheter biofilm suggesting that ethanol treatment specifically targets *Candida* biofilm formation. As inhibitory effects of ethanol concentrations of 10%, 20%, and 80% were evaluated, similar reduction rate in dry biofilm weight and biomass was seen (Mukherjee *et al.*, 2006). This result indicates that ethanol concentration as low as 10% could be equally effective in inhibiting biofilms in *C. albicans*. Even though it has been seen that ethanol treatment can reduce the biofilm activity of *C. albicans*, further research regarding its effectiveness against biofilms of other microorganisms in a polymicrobial model, different concentrations, and various catheter material types is necessary.

#### Recent treatments: chelating agents and biofilm dispersants

Chelating agents may destabilize the biofilm structure, and some of them shown to have antimicrobial properties against bacteria and fungi. Venkatesh *et al.* (2009) looked at the

synergistic application of catheter lock solutions of Ethylenediaminetetraacetic acid (EDTA), *N-acetylcysteine* (NAC), talactoferrin (TLF), and ethanol alone or in combination with antibiotics. The study showed chelating agent combination with antibiotics was effective against biofilms of *C. albicans* and *Staphylococcus epidermidis* in catheters. These agents are originally known to have various inhibitory effects that allowed them to synergistically act as antibiofilms. EDTA inhibits planktonic *Candida* and *Staphylococcus* species, NAC disrupts EPS formation, and TLF has antimicrobial activity (Venkatesh *et al.*, 2009). All 8 mg/mL of EDTA, NCA, TLF and 12.5% of ethanol decreased both the mean biofilm mass and thickness in monomicrobial and polymicrobial *C. albicans* ATCC 32354 biofilm (Venkatesh *et al.*, 2009). Among all the above agents, TLF was the least effective, while ethanol was more successful in reducing monomicrobial and polymicrobial biofilms of *C. albicans* (Venkatesh *et al.*, 2009). Using chelating agents resulted in substantial change in biofilm structure suggesting that this treatment could potentially be used to eradicate *C. albicans* biofilms from catheters.

Another treatment strategy to reduce biofilms in this organism is dispersal and shedding of the daughter cells. A range of factors have shown to induce biofilm dispersal across microorganisms. Various harsh physical environments, limited nutrient availability, and quorum-sensing (QS) can cause shedding of cells from biofilms. At the same time, it is not entirely understood whether the detachment occurs due to a controlled biological process or some environmental stimuli. It is suggested that biofilm dispersion could assist in disrupting the *C. albicans* biofilm resulting in planktonic cells that could later be targeted with antifungals to be removed from catheters. While this phenomenon needs further research in *C. albicans*, Thormann *et al.* (2003) stated that detachment in some microorganisms such as *Shewanella oneidensis* MR-1, *Acinobacter spp.*, and *Pseudomonas spp.* was triggered as a result of nutrient

starvation or electron source disruption (Barraud *et al.*, 2006). Another QS disruptor depend on the chelating agent, EDTA which was observed to cause cell lysis, loss of cell viability, and increase in cell sensitivity (Banin *et al.*, 2006). EDTA not only dispersed the *Pseudomonas aeruginosa*, it also killed *P. aeruginosa* biofilms (Banin *et al.*, 2006).

The organisms in the biofilm use QS signaling to grow and increase their pathogenicity. Inhibition of their QS by dispersing the daughter cells in the biofilm needs through research due to biofilm's heterogeneity. Currently, few research studies looked at the QS mechanism on sloughing off biofilms of *C. albicans*, but studying the behavior of other microorganisms could provide us with hints to solve this infection. Rice *et al.* (2005), indicated that nutrient-rich condition induced a QS-dependent detachment in an opportunistic pathogen, *Serratia marcescens*, biofilms. Some studies characterized a number of QS disruptors such as *cis*-2-decanoic acid (DCA), diffusible signal factor (DSF), and farnesol that inhibited the germ tube formation (mycelia) by *C. albicans* respectively in the order of most effective to least effective (Boon *et al.*, 2008). In order to understand the mechanism relating to these unsaturated fatty acids, it is important to note that typically *C. albicans* exist in yeast form, but they can also form germ tubes that are able to enter bloodstream in humans and cause infections (Wang *et al.*, 2004). DCA and DSF have very similar structures verified by HPLC and NMR, and DSF is known to be a QS signal working in cell communication in a range of bacterial species (Boon *et al.*, 2008). DCA significantly decreased the hyphal structure of *C. albicans* by 15% (Boon *et al.*, 2008).

Although the molecular mechanisms of DCA needs more work, the DCA ability to target cross-kingdom interactions may lead us one step ahead in treating biofilm infections in *C. albicans*. Similarly, Davies & Marques (2009) identified that DCA, made by *P. aeruginosa*,

could disperse biofilms of *C. albicans* in vitro as well as biofilms formed by *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Streptococcus pyogenes*, *Bacillus subtilis*, and *Staphylococcus aureus*. It was suggested that application of DCA degraded the EPS of biofilms which could then be followed by antimicrobial agents to kill the floating daughter cells (Davies & Marques, 2009). Cell-cell communication signals seem to be one of the promising strategies to target when trying to control infectious diseases. The active detachment of cells from their biofilm nature is helpful in decreasing the BSIs, but the triggering factors reaching this nature is not completely understood and needs to be researched. After biofilm dispersal, further treatment with antimicrobial agent could remove the biofilms and effectively eliminate biofilm formation in indwelling device, prevent growth of organisms on the catheter, and improve patient's lives.

### **Conclusion and Outlook**

Intravascular catheters are widely used in medicine, allowing for administration of intravenous fluids, blood, medications, and nutrition. However, their use is associated with a high risk of BSIs caused by colonization of microorganisms. One of these organisms, *Candida albicans*, has shown to be one of the leading organisms responsible for these infections by forming biofilms. Polymicrobial biofilms of *Candida* species add to the complexity of the biofilm which complicates the biofilm prevention solution. *C. albicans* is a commensal organism possessing characteristics that allows it to survive harsh environments and to be flexible in changing from harmless commensal to invasive pathogens. This organism form highly resistant biofilms in catheters increasing its pathogenicity which has been difficult to target with just antimicrobials and antifungals. This low success in treatment of these infections using antimicrobials urges us to seek alternative treatment strategies.

On the basis of the available studies, ALT, chelating agents, ethanol, and QS disruptors appear to be the most promising prevention strategies for catheter-related BSIs. Literature on BSIs has shown that application of ethanol in ALT has been the most effective method in preventing BSIs. On a proteomic level, Mukherjee et al. (2006) reported that disruption of the protein, *adh1p*, that produces ethanol prevented biofilm formation in *C. albicans* through an ethanol-dependent pathway. Additionally, chelating agents may destabilize the biofilm structure, and some of them have been shown to have antimicrobial properties against bacteria and fungi, but caution must be exercised that these agents do not lead to resistance/tolerance. However, current efforts lack to present a combined ALT approach with ethanol and a chelating agent that is both effective and not harmful to the human body upon administration.

Despite the high effectiveness of the ALT, there are a number of concerns with the approach. Sufficient care is needed when setting an antimicrobial lock method since a leak in the catheter could flush toxic concentrations of the antibiotic agent in the systemic circulation of the patients leading to toxicity and possibly death. Another concern is that flushing high concentrations of strong antimicrobial agents could lead to antimicrobial resistance. Considering these concerns, ethanol could be the most optimal lock solution because of its antimicrobial activity against a broad spectrum of microorganisms, its low cost, and its absence report of tolerance.

Despite the high number of incidence, these infections are preventable and effective strategies are needed to make progress toward the goal of eliminating BSIs. There is a great need to provide an optimal strategy to prevent catheter-related bloodstream infections associated with *C. albicans*. After all, development of a novel mechanism that could remove the biofilms and



effectively eliminate biofilm formation in indwelling device, prevent growth of organisms on the catheter, and improve patient's lives is necessary.

Table 1. Factors affecting *C. albicans* biofilm to disperse.

<b>Factor</b>	<b>Organisms</b>	<b>References</b>
Nutrient starvation	<i>Shewanella oneidensis</i> MR-1, <i>Acinobacter</i> spp., and <i>Pseudomonas</i> spp.	Barraud <i>et al.</i> , 2006
Nutrient rich	<i>Serratia marcescens</i>	Rice <i>et al.</i> , 2005
Electron source disruption	<i>Shewanella oneidensis</i> MR-1, <i>Acinobacter</i> spp., and <i>Pseudomonas</i> spp.	Barraud <i>et al.</i> , 2006
QS	<i>Shewanella oneidensis</i> MR-1, <i>Acinobacter</i> spp., and <i>Pseudomonas</i> spp., <i>Candida albicans</i>	Barraud <i>et al.</i> , 2006 Rice <i>et al.</i> 2005,
EDTA	<i>Pseudomonas aeruginosa</i>	Banin <i>et al.</i> , 2006
Unsaturated fatty acids: <i>cis</i> -2-decanoic acid (DCA)	<i>Candida albicans</i> <i>Escherichia coli</i> , <i>Klebsiella pneumonia</i> , <i>Proteus mirabilis</i> , <i>Streptococcus pyogenes</i> , <i>Bacillus subtilis</i> , and <i>Staphylococcus aureus</i>	Boon <i>et al.</i> , 2008; Davies & Marques, 2009
Unsaturated fatty acids: diffusible signal factor (DSF)	<i>Candida albicans</i> , <i>Pseudomonas aeruginosa</i>	Boon <i>et al.</i> , 2008; Wang <i>et al.</i> , 2004; Davies & Marques, 2009
Unsaturated fatty acids: farnesol	<i>Candida albicans</i>	Boon <i>et al.</i> , 2008

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