

## Evaluation of bacterial contamination arteriovenous fistula by direct contact for hemodialysis patients in Zawia Kidney Center

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Abstract:		

Dialysis is the process of removing toxins directly from the blood i.e. hemodialysis or indirectly i.e. peritoneal dialysis using diffusion across a semipermeable membrane. Chronic care patients undergoing hemodialysis for treatment of end-stage renal failure experience higher rates of healthcare-associated infection, in this study, our objective is to assess the level of contamination that reaches hemodialysis patients via the arteriovenous fistula. Hemodialysis patients are always at high risk of infection due to the nature of the hemodialysis procedure which exposes the bloodstream to the external environment, which opens up ways for bacteria, viruses, and fungi to enter the human body and cause contamination, which ultimately causes the infection. The contamination can occur from any part of the dialysis procedure, whether it is from the water being used, the machinery used, the catheter, or through direct contact with the medical staff. Most medical procedures for hemodialysis help decrease the possibility of contamination, however, the possibility of contaminations will always remain and should be monitored and regulated, which is why it is necessary to determine the cause of these contaminations and what parameters affect it.

Keywords: bacterial contamination, arteriovenous fistula, Zawia kidney center.

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# تقييم التلوث الجرثومي للناسور الشرياني الوريدي عن طريق الاتصال المباشر لمرضى الغسيل الكلوي في مركز الكلى الزاوية

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لملخص:

غسيل الكلى هو عملية إز الة السموم مباشرة من الدم، أي غسيل دموي أو بشكل غير مباشر، أي غسيل الكلى البريتوني باستخدام الانتشار عبر غشاء نصف منفذ. يعاني مرضى الرعاية المزمنة الذين يخضعون لغسيل الكلى لعلاج الفشل الكلوي في المرحلة النهائية من معدلات مرتفعة من العدوى المرتبطة بالرعاية الصحية، ويكون مرضى غسيل الكلى دائمًا معرضين لخطر الإصابة بالعدوى بسبب طبيعة إجراء غسيل الكلى الذي يعرض مجرى الدم للبيئة الخارجية، والتي يفتح طرقًا لدخول البكتيريا والفيروسات والفطريات إلى جسم الإنسان وتسبب التلوث، مما يؤدي في النهاية إلى الإصابة بالعدوى. يمكن أن يحدث التلوث من أي جزء من عملية غسيل الكلى، سواء كان ذلك من المياه المستخدمة، الألات المستخدمة، القسطرة أو من خلال الاتصال المباشر من الطاقم الطبي. تساعد معظم الإجراءات الطبية لغسيل الكلى على تقليل احتمالية التلوث، ومع ذلك، فإن احتمالية التلوث ستظل قائمة دائمًا ويجب مر اقبتها وتنظيمها، ولهذا السبب من الضروري تحديد سبب هذه التلوثات وما هي العوامل التي تؤثر عليها.

### الكلمات المفتاحية: التلوث الجرثومي، الناسور الشرياني الوريدي، مركز الكلى الزاوية.

#### 1. Introduction

In the event of kidney failure or 85% to 90% of the kidney function is lost i.e. the glomerular filtration rate (GFR) is less than 15%, dialysis may be instituted to substitute the function of the kidney [1][2]. Dialysis is the separation of smaller molecules from larger molecules in solution by diffusion of the small molecules. Dialysis removes metabolic waste products as well as excess water and sodium, which contributes to regulating blood pressure, and maintains the levels of many chemicals within the body [3]. Majority of signs and symptoms of renal failure can be relieved by dialysis and there are generally to methods of dialysis, continuous ambulatory peritoneal dialysis (CAPD) and hemodialysis.

For continuous ambulatory peritoneal dialysis, the peritoneal membrane that lines the abdominal cavity becomes the dialyzing membrane and one to two liters of sterile glucose/salt solution is injected into the abdominal cavity [4]. Small molecules such as potassium and urea then diffuse into this solution, which is then drained and discarded. In CAPD dialysis, this process is usually done several times every day. As for hemodialysis, the patient's blood is pumped through a machine that acts as an artificial kidney. The blood is then separated from a balanced salt solution by a membrane that is cellophane like and small molecules start to diffuse across the membrane. As for the excess fluids, they are removed by applying pressure to the blood and the filtering it [5]. Hemodialysis is usually done three times a week (4 to 6 hours per session) in a medical facility or at home, which makes far more efficient them CAPD dialysis in terms of removing wastes.

As it is clear from the description of both methods, treating chronic kidney disease via dialysis requires constant treatment and caution. Another issue with dialysis is the fact that external assistance is required for it to be administered and the inside of the body needs to be interacted with, whether by injecting into the body or transferring blood from and to the body, which can lead to complications or side effects that may harm or endanger the patient. Since dialysis is crucial for the life of patients that have chronic kidney disease, understanding and thereby minimizing or avoiding these complications and side effects is of great benefit and should be the subject of extensive research. One of these complications is an extremely common in dialysis patients, which is infection caused by contamination, and it will be the focus of this study.

The study of contamination due to hemodialysis is a topic that has been studied meticulously due to its serious effect on the patients' health. Many different studies have been conducted in order to determine the causes of contamination, whether it is the bacterial or viral cause of the contamination or the source of said bacteria or virus. Some studies have chosen to focus on the water used in dialysis centers and examples of such studies include a study by Montanari et al, in which the water from the water system used in a hemodialysis center in Brazil is tested for contamination. 182 samples were taken from the water distribution system, kidney machine water and reuse system and the results of the study showed that 32 samples were Gram-positive rods, 120 Gram-negative rods, 20 Gram-positive cocci and 11 mycobacteria [6]. Another study that focused on the water system in a hemodialysis center was a study conducted in Iraq by Humudat et al. in this study, bacteriological characteristics and endotoxin contamination levels in hemodialysis water produced in dialysis centers are examined and evaluated. Forty-eight water samples were collected and analyzed from four major hospitals one year to evaluate seasonal effects. Results showed that 44% of the tested samples for bacterial levels are higher than the maximum value set out by the international standards and 44% of the endotoxin values exceeded the international standards [7]. A third similar study was conducted in Nigeria by Okunola et al [8], where water samples were aseptically and serially collected from three hemodialysis units. The samples were taken from six points at three center coded over a 6-month period. Results have shown that the water system in all three of the dialysis centers were contaminated with 13 Gram-negative aerobic bacteria such as Pseudomonas species and Moraxella species at all the points in the three centers of dialysis wards and contamination of hemodialysis devices to determine the source of bacteria for infections. All bacterial samples were collected by the swab method and the agarose stamp method the bacteria were identified by BBL CRYSTAL Kit or 16s rRNA sequences. Results show that bacterial cell number of hemodialysis device was lower than environment of patient surrounds. However, Staphylococcus spp. was found predominantly on the hemodialysis device (46.8%). Among Staphylococcus spp., Staphylococcus epidermidis was most frequently observed (42.1% of Staphylococcus spp.). A study by Villanueva et al. [9] chose to focus on the contamination caused by cross-transmission, which used structural analysis to analyze the results of a simulated epidemic model. The simulation was carried out in a Dialysis Unit equipped with 19 machines for 62 patients. One of these patients was randomly chosen and considered as a carrier of an infectious agent capable of being transmitted to other patients, by means of the shared use of the same dialysis machine. Results have shown that in 10 days, 87.09% of patients could have been exposed to the infection, and 68.42% of the machines could have been contaminated. Some studies focused on the contamination that could be caused by the staff, such as the study by Alfurayh et al. [10], which used a clinic virological study in hemodialysis centers to investigate if the hands of dialysis personnel could represent a mode of transmission of Hepatitis C Virus (HCV) among

patients. In this study, one liter of sterile water was used for each hand washing of dialysis personnel and was collected in a sterile container and tested for HCV-RNA by polymerase chain reaction (PCR) within 3h of collection. Eighty hand washings from nurses dialyzing HCV-positive patients and 100 hand washing from nurses dialyzing patients) were tested for HCV-RNA. As a control, 60 hand washings were collected from the dialysis personnel before entering the dialysis unit and tested for HCV-RNA. Results show that HCV-RNA was positive in 23.75% of samples of group A, in 8% of samples of group B and in 3.3% of samples of group C. the study concluded that despite the adherence to the standard precautions, the presence of HCV-RNA on the hands of some dialysis personnel was still detected. As for studies in Libya, there are not many studies conducted on the topic of contamination in hemodialysis. Example of such studies include a study by Shahlol et al [11], whom studied the bacterial biofilm that contaminated indwelling catheter tips for renal dialysis machines among patients and the associated resistance of antibiotics in Brack Hospital. One hundred of catheter tips were collected from patients. Two inches of the distal tip of the catheter were clipped directly into a sterile broth. The culture was by rolling the tip across blood agar and incubated aerobically at 37C° for 18-24 hours. Results show that the Bacterial isolates were Staphylococcus aureus 33%, Bacillus spp. 9.7%, Klebsiella pneumonia 8.3%, Escherichia coli 8.3%, Staphylococcus albus 8.3%, Enterobacter aerogenes 8.3%, Enterobacter spp. 6.9%, Proteus mirabilis 6.9%, Klebsiella ozaenae 6.9%, and Acinetobacter 2.7%. Another study by Alfarisi et al.[12] aimed to determine the physicochemical and bacteriological characteristics of water used by hemodialysis services and to isolate and identify fungi present in water systems of hemodialysis units in Benghazi Medical center. Results show that from the six samples that have undergone chemical analysis and 32 samples for microbiological test, Chemical parameters of study area A and B for drinking and dialysis water were within international standards, but there was a minute of Calcium elevation in area B that was 3.3mg/l. No contamination with bacteria observed in all samples in section (A), the counts of yeasts and 49 filamentous fungi investigated in the tap water, in the treated water, and dialysis machine in section B and Penicillium spp was the most frequent fungi. Based on the review of the literature, it is clear that there are not many studies the focus on the contamination caused by contact and even less studies have been conducted on contamination in Libya. This makes this study even more important since it covers a topic that is rarely covered by other literature.

#### 2. Materials and methods

This study was conducted from was conducted among 100 patients at Zawia kidney center from October first to November fifteenth 2022 direct contact between the patient and medical staff at Zawia kidney center the swab samples were collected from the location of the using sterile cotton swab under an aseptic condition, then specimens were transferred into sterile nutrient broth in screw caped test tubes and were delivered to the laboratory within 1hr. The arteriovenous fistula swabs inoculated thioglycate broth and incubated for

24hrs at  $37C^{\circ}$  to allow for bacterial growth. Once the 24hr incubation period is over, the samples are placed in different mediums for 24 hours at 37 C° to determine which bacteria exist in the sample, these mediums include MacConkey agar, blood agar Chocolate Agar (CAP or CHOC), Sabourau Dextrose agar and mannitol agar media then the medical staff at Zawia kidney center samples were incubated at 37 °C for 24hrs. Data were then recorded and analyzed, and the results were presented as graphs using Microsoft Excel.

**3. Result:** - Based on the data results from the laboratory tests that were conducted, there were 12 samples that were contaminated out of the 86 total samples. This means that there was a contamination rate of 13.63%. Test results also show that the causes of these contaminations are two types of bacteria, which were identified as Klebsiella and Staphylococcus Hemolytic.

For the breakdown of each age group by gender, the data shows that the highest number of contaminated samples was for the age group between 40 - 59 years old for both male and female patients. Out of the 33 male samples in this age group, five of them had contamination, which amounts to a 15.15% contamination rate, while the female samples had two out of the nine samples, which gives a contamination rate of 10.53%. As for the other age groups, the age group of 1-17 years old had no contaminated samples (there were no female samples in this age group) and the female sample for age group 60-80 years old had a contamination rate of 14.29%. As for the men sample both age groups 18-39 and 60-80 had a contamination rate of 18.18% due to having both the same number of total samples and number of contaminated samples.



Figure 1: distribution of Staphylococcus and Klebsiella bacteria in different age groups.

The data reveals that the highest number of contaminated samples was found among individuals aged 40 to 59, regardless of gender. Among the male patients in this age group, five out of 33 samples were contaminated, resulting in a contamination rate of 15.15%. Among the female patients in the same age group, two out of nine samples were contaminated, resulting in a contamination rate of 10.53%. In the other age groups, there were no contaminated samples among the 1-17-year-old group (with no female samples in this group). Among female patients aged 60-80 years, one out of seven samples was contaminated, resulting in a contamination rate of 14.29%. For male patients, both the 18-39 and 60-80 age groups had an identical contamination rate of 18.18%, as they had the same number of total samples and the same number of contaminated samples.



Figure 2: distribution of Klabsila and S.coccous Hemolytic with age groups.

The distribution of bacteria in the samples was examined, and it was observed that Staphylococcus was present in all age groups except for the 1-17 years old group, which did not exhibit any contamination. Furthermore, Staphylococcus Hemolytic was identified in all age groups, suggesting its presence across the entire range. However, Klebsiella was only detected in the 40-59 years old age group.



Figure 3: duration of dialysis and the occurrence of contamination with gender.

Samples with a dialysis duration of 20 years or more showed no evidence of contamination, indicating that patients undergoing dialysis for 20 years or longer had uncontaminated samples. Analyzing the data by gender, contamination was observed exclusively in female samples with dialysis duration of five years or less. In contrast, among the male samples, the number of contaminated samples decreased as the duration of dialysis increased, except for those who had undergone dialysis for 20 years or more, as their samples showed no signs of contamination.



Figure 4: the different bacteria in relation to the duration of dialysis.

The results indicate that Klebsiella was exclusively detected in samples from patients with a dialysis period of 5 years or less. On the other hand, Staphylococcus Hemolytic was found in all samples regardless of the duration of dialysis.

#### 4. Discussion

T The results indicate that the overall contamination rate of the sample was 13.63%, which is relatively high considering that patients undergoing dialysis can have weakened immunity, increasing the risk of infection. Although a contamination rate of 13.63% may appear lower than the contamination rates reported in the referenced studies in the literature review, such as the study conducted by Humudat et al. with a contamination rate of 44% [7], it is important to note that those studies focused on equipment and water supply contamination, which is more challenging to control compared to contamination resulting from direct contact. While it is not expected that all cases of contamination will lead to infection, every instance of contamination should be treated as a potential infection case that could have serious implications for the patients' health. Therefore, a 13.63% contamination rate resulting from contact is high and unacceptable; particularly considering that adherence to basic medical procedures should either eliminate or significantly reduce the likelihood of such contamination. He results also revealed that the cause of contamination was attributed to two types of bacteria, namely Klebsiella and Staphylococcus Hemolytic, with the latter exhibiting significantly higher contamination rates than the former. The predominance of Staphylococcus as the primary cause of contamination aligns with other studies, such as the study by Shimohata et al. [13], which reported that 47% of the contaminated samples had Staphylococcus, as well as the study by Danese et al. [14], which found that 54% of the samples had Staphylococcus, along with other studies [15], [16]. Klebsiella, although present in other contamination studies, occurred at much lower rates [8], [16].

When examining the breakdown of results by gender, it was observed that males had a higher contamination rate compared to females. While part of this difference could be attributed to the fact that there were more willing male participants in the study than females, it is also possible that the predominance of female medical staff influenced the results. This could be because female staff may feel more comfortable working with female patients, resulting in greater attention to medical procedures compared to male patients. However, it should be noted that the sample size and scope of this study are not sufficient to determine whether the gender difference can increase the risk of contamination, but it does indicate the presence of a correlation.

Similarly, age group and dialysis period of the patients demonstrated a correlation with the contamination rate, particularly the dialysis period, where the contamination rate decreased as the patient's dialysis period increased. This trend could be attributed to the fact that patients undergoing dialysis for longer periods become more familiar with the dialysis procedure, leading to improved adherence to infection control measures

#### 5. Conclusion

The overall contamination rate for all samples was 13.63%, which is alarmingly high for dialysis patients who are already at risk. The primary causes of contamination were identified as two types of bacteria, namely Klebsiella and Staphylococcus Hemolytic. Among them, Staphylococcus Hemolytic was found to be the most prevalent and the Male patients were found to be three times more likely to be at risk of contamination compared to female patients. Additionally, patients in the age group of 40-59 exhibited the highest contamination rates, although not significantly higher than other age groups, except for the age group of 1-17, which showed no contamination rates. Interestingly, the longer a patient had undergone dialysis, the lower their likelihood of experiencing contamination. Based on these findings, it is crucial to implement effective measures to reduce contamination rates in dialysis centers.

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