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Research Article

Evaluating the Effectiveness of Various Detergents and Disinfectants on Bacteria Isolated from Newborn Incubators in Two Major Hospitals in Misurata, Libya

Abubaker Elrotob[1*], Nabil Al-Atrash[2], Khawla Al-Darwish[2], Mastoura Ben Saeed[2]

[1]*Department of Epidemiology, Health Science Faculty, Misurata University, Libya*

[2]*Department of Microbiology, Science Faculty, Misurata University, Libya*

**Corresponding author: elrotobbaba@gmail.com*

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ABSTRACT

Hospital infections are considered to be a major contributor to the global spread of many diseases, especially when it comes to the spread of antibiotic-resistant bacteria. This study evaluates the effectiveness of sterilizers and disinfectants, including Minuson AF, ethanol, propanol AF, and popular sodium hypochlorite, as well as two alternative options, commercial sodium hypochlorite (Clorox) and Dettol. The assessment is conducted against bacteria isolated from swabs taken from newborn incubators in the public Misurata Medical Center and the private Al-Saeed Hospital, both significant healthcare facilities in Misurata City in Libya. The antimicrobial activity of disinfectants was evaluated using the disc-diffusion method on Mueller-Hinton agar-filled petri dishes containing bacterial suspensions. They underwent differential and biochemical testing, and bacterial species were isolated from the samples that underwent testing. A total of 164 samples 145 positive samples were obtained, with 87 (60%) coming from Misurata Medical Center and 58 (40%) coming from Al-Saeed Private Hospital. The isolation of various bacterial strains, including *Staphylococcus aureus* (57.24%), *Bacillus* spp (23.45%), *Klebsiella* spp (7.59%), *Streptococcus pneumoniae* (4.13%), *Pseudomonas spp* (3.45%), *Staphylococcus epidermidis* (2.07%) and *Escherichia coli* (2.07%), revealed the inefficacy of sterilization in certain incubators across both hospitals. The results indicated that certain disinfectants and sterilizers, such as Minuson AF, Clorox, and Dettol, were effective against all isolated species. Ethanol demonstrated effectiveness against all species except *Klebsiella* spp. Propanol AF showed effectiveness against *Bacillus* spp and *Pseudomonas spp*, while popular sodium hypochlorite (PSH) was effective against all species except *S. epidermidis* and *Klebsiella* spp. Moreover, combining two of these substances was found to enhance the efficacy of some while diminishing the efficacy of others.

Keywords: Hospitals, Newborn incubators, Isolated bacteria, Detergents and disinfectants, Antibacterial efficacy.w

INTRODUCTION

An infection that strikes a patient in a hospital or other healthcare facility is known as a nosocomial infection [1]. A nosocomial infection is any infection that the patient contracts after being admitted to the hospital (i.e., the patient did not have the infection when he was admitted) and does not manifest for at least 72 hours following admission [2]. Numerous factors contribute to this infection, including the patient's immunity and personal hygiene; hospital environment factors, such as the level of sterilization and treatment efficiency; medical staff factors, such as the extent of precautions taken; etc. Hospital infections have been rising annually from 5 to 10% in developed nations like America and Europe to more than 40% in some regions of Asia [3]. Hospital surfaces contaminated with pathogenic bacteria are significant sources of healthcare-associated infections (HCAIs). Regular assessment of the effectiveness of disinfectants used for cleaning hospital surfaces is crucial for HCAI prevention and control. However, many resource-constrained nations lack routine evaluation of disinfectant effectiveness [4]. Exogenous acquisition of infections occurs when individuals come into contact with contaminated surfaces or other individuals, including caregivers, patients, or healthcare workers [5]. In the United States, over 2 million nosocomial infections have been attributed to antibiotic-resistant bacteria [3]. Hospital infection rates are a major contributing factor to the global increase in disease and death rates. An estimated 9,600 to 20,000 patients experience bacteremia-related deaths each year in the United States, where there are over 2 million hospital infections among adults and infants. Of these infections, 50 to 60% are caused by antibiotic-resistant organisms. On the other hand, newborns with hospital infections have longer hospital stays, more expensive care, and a 50% mortality rate by the time they are two weeks old [6].

According to the National Surveillance System (NNIS) in the United States, rates of nosocomial infections (NI) in neonatal intensive care units (NICUs) range from 1.8% to 39.8%. Globally, NI rates in NICUs vary between 8.4 and 57.7% across different countries [7]. Additionally, pre-term newborns in NICUs come into close contact with their surroundings, which includes the medical equipment and incubator they are housed in, as well as with their parents and other caregivers, all of whom have the potential to harbor bacteria [8]. The study by Hoseini *et al.* revealed that out of 3,129 patients admitted to three hospitals' neonatal intensive care units (NICUs), 208 were diagnosed with nosocomial infections. Premature and low birth weight infants are particularly vulnerable to these infections. This underscores the critical importance of maintaining vigilant efforts to prevent and manage nosocomial infections in NICUs [9]. Furthermore, neonatal mortality rates (NMR) in low- and middle-income countries (LMICs) are alarmingly high, ranging from 40 to 50 per 1000 live births, with infections being the primary cause. Preterm neonates, especially those in NICUs, face an elevated risk of contracting nosocomial infections (NIs) due to their immature immune systems. To address this challenge, NICUs should adopt standardized cleaning protocols and regularly conduct environmental sampling to reduce hospital surface contamination, minimize the adverse effects of NIs, and ensure appropriate antibiotic usage [10]. A notable level of bacterial contamination was observed on objects and instruments within the NICU, posing a significant risk of nosocomial infections [11]. Common potential pathogens isolated include *E. coli*, *Klebsiella* spp, and *S. aureus*. According to Wesam Hassanein *et al.*, prevalent causative agents of healthcare-associated infections include *E. coli*, *S. aureus*, *Klebsiella spp*., and Coagulase-negative Staphylococci (CoNS) [2]. Additionally, Asinobi *et al.* found that the most common isolates in their study were *S. aureus*, Coagulase-negative Staphylococcus, and *E. coli*, emphasizing the need for improved hygiene standards among healthcare providers to mitigate the burden of nosocomial infections [12]. Various bacteria, including *S. aureus*, *Streptococcus spp*., *B. cereus*, *Acinetobacter spp*., *Enterococci*, *P. aeruginosa*, Legionella, and members of the Enterobacteriaceae family such as *E. coli*, *P. mirabilis*, *Salmonella spp*., *S. marcescens*, and *K. pneumoniae*, are frequent causes of nosocomial infections. However, *E. coli*, *S. aureus*, *Enterococci*, and *P. aeruginosa* are most frequently reported as nosocomial pathogens [3].

In hospitals and other healthcare settings, antiseptics and disinfectants play a crucial role in preventing nosocomial infections and are integral to infection control protocols [13]. The majority of bacteria resistant to multiple drugs also show resistance to commonly used disinfectants, primarily due to the presence of comparable mobile genetic elements (MGEs), particularly plasmids carrying genes encoding resistance to both antibiotics and disinfectants [13,14]. The spread of multidrug-resistant (MDR) bacteria poses a significant challenge in healthcare environments, complicating patient treatment and leading to worse outcomes [15]. MDR, characterized by resistance to at least one substance from three or more antimicrobial classes, is a growing global concern and is estimated to cost US$100 trillion annually, resulting in 10 million deaths by 2050 [16,17]. In a study conducted by Reboux *et al.*, the bacteria identified showed resistance to disinfection methods and encompassed Coagulase-negative Staphylococci, *Enterococcus*, *Enterobacteria*, and *Bacillus* strains [18]. According to Alharbi's study, there is microbial contamination in newborn incubators, which presents problems for this susceptible population. Bacterial growth in blood cultures was found to be diverse. Coagulase-negative *Staphylococcus* was the most commonly isolated species, followed by *Klebsiella* spp., *S. agalactiae*, *E. cloacae*, *E. coli*, *A. baumanii*, and *Candida* spp. Furthermore, methicillin-resistant *S. aureus*, *P. aeruginosa*, and *Bacillus* spp. were identified from single isolates [19]. An increased frequency of antibiotic-resistant gram-negative bacteria was found in sepsis patients in the neonatal intensive care unit and neonatology unit, according to a study by Silago *et al.* Additionally, it is extremely alarming that a high prevalence of multi-drug resistant gram-negative bacteria (MDR-GNB) is colonizing infants, contaminating the mothers' hands, and contaminating the infants' environment [17].

Healthcare facilities rely heavily on disinfectants as part of their infection control strategies to prevent the spread of healthcare-associated infections (HCAIs). Chemical agents are employed to eliminate bacterial, viral, and fungal contaminants from hospital surfaces, including patient beds, side tables, trolleys, and benches. This process substantially lowers the likelihood of healthcare-associated infections (HCAIs) [20]. According to the study conducted by Elrotob *et al.*, certain disinfectants were found to be ineffective, particularly when used during the initial stages. The study underscored the significance of evaluating the quality, as well as the storage and handling conditions, of these disinfectants and sterilizers.[21] Furthermore, freshly made 0.5% sodium dichloroisocyanurate is more effective and potent against standard strains and clinical isolates than 4.8% chloroxylenol. But when it is kept in storage for more than 48 hours, especially for clinical isolates, its effectiveness dramatically diminishes [4]. The mechanism of action of disinfectants and antiseptics is still not fully understood, especially when it comes to how they affect different types of infectious agents besides bacteria. Although research on bacteria has advanced significantly, a greater understanding of these mechanisms in other pathogens is obviously needed. Additionally, the choice of antiseptic or disinfectant product should be customized to specific conditions or nosocomial outbreaks [13].

Hence, our study concentrates on evaluating several disinfectants and sterilizers against bacteria isolated from children's incubators in the intensive care units of the two mentioned health facilities. Additionally, we examine the impact of combining certain disinfectants with each other. This decision stems from our observation during swab collection, noting the sequential use of popular sodium hypochlorite (PSH) for cleaning, followed immediately by sterilization or disinfection without any interval between them.

MATERIAL AND METHOD

Collection, transportation of specimens, and isolation of bacteria

In the public Misurata Medical Center and the private Al-Saeed Hospital, two major healthcare facilities in Misurata, a total of 164 samples were obtained from newborn incubators. A comprehensive swabbing of the entire nursery was conducted, with isolates collected from various areas of the incubators, including feeding openings, ventilation openings, front openings, and the entire incubator. Each isolate was assigned a unique identifier, and pertinent details such as the date, sample location, and the presence or absence of a child in the nursery were recorded. Following swabbing, samples were collected using a sterile saline solution. The swabs were then inoculated into nutrient broth tubes, followed by a 24-hour incubation period at 37°C. Subsequently, samples were streaked onto blood agar (Merck Germany), Nutrient agar (Oxoid, UK), and MacConkey agar (Merck Germany) plates using a sterile loop. After incubation, all isolates underwent characterization using standard protocols, which included evaluating colony morphology, culture characteristics, gram staining, and biochemical reactions.

Preparation of bacterial suspensions

Briefly, bacterial suspensions equivalent to 0.5 McFarland turbidity standard solution were prepared by subculturing one isolated colony from blood agar or MacConkey agar into sterile 0.85% physiological saline. These suspensions were then swabbed onto entire plates of Mueller-Hinton agar (MHA) (Oxoid, UK).

Antibacterial activity of disinfectants and detergents on isolated bacteria

The disc-diffusion method was employed on Petri dishes containing Mueller-Hinton agar, utilizing 6 mm filter paper discs.[12,21] Six types of disinfectants and detergents were tested, including four commonly used in hospitals: Minuson AF, Ethanol, Propanol AF, and Popular sodium hypochlorite. Additionally, two alternative agents were included: Commercial sodium hypochlorite (Clorox) and Dettol (refer to Table 1). Combinations of propanol AF and Minuson AF, Propanol AF and Ethanol, Minuson AF and Ethanol, as well as Minuson AF and popular sodium hypochlorite (PSH) were also evaluated. The bacterial suspension was cultured onto Mueller-Hinton agar medium using a cotton swab, and the plates were left in a sterile environment at room temperature for 10 to 15 minutes to allow the surface of the medium to dry. Filter paper discs were then saturated with 50 μ (microliters) of each disinfectant or compound using a pipette. With a sterile metal needle, the discs were distributed onto each plate, ensuring space between them to distinguish the zones of inhibition. On the first plate, four discs were placed containing Minuson AF, ethanol, propanol AF, and popular sodium hypochlorite. In the second plate, combinations of two disinfectants were tested (25 + 25 μL each), including (Popular hypochlorite sodium (PSH)& Minuson AF), (Minuson AF & Propanol AF), (Propanol AF & Ethanol), and (Minuson AF & Ethanol). Similarly, commercial Clorox and Dettol were tested individually on these isolates to explore additional alternative options for sterilization, following the same procedure. After incubating the plates at 37°C for 24 hours, the sensitivity effect of these materials was assessed based on the area of inhibition surrounding the saturated disc, categorizing them into sensitive, moderately sensitive, and resistant, as depicted in Fig. 4.

Data analysis and visualization, including the creation of tables and figures, were conducted using Excel 2016.

Table 1: displays the disinfectants and detergents utilized, along with their respective manufacturers

Disinfectant/Sterilizer	*Concentration*	*Place of manufacture*
Ethanol	70%	China
Propanol AF	70%	Turkey
Popular sodium hypochlorite (PSH)	10%	Libya
Minuson AF	60%	Poland
Dettol	3%	Germany
Commercial Sodium hypochlorite (Clorox)	10%	Turkey

RESULTS

Based on the findings from the entire set of samples of 164 swabs from two sizable hospitals in Misurata, Libya, the Misurata Medical Center, a public facility, had 101 swabs (62%), and Al-Saeed Private Hospital had 63 swabs (38%), as shown in Fig. 1. The swabs taken from the newborn incubators in both facilities revealed bacterial contamination within the incubators.

Out of 164 samples, bacterial growth was observed in 124, resulting in a total of 145 bacterial isolates retrieved from these samples, indicating the presence of bacterial growth. Of these, 87 samples (60%) came from the public hospital (Misrata Medical Center) and 58 samples (40%) from the private hospital Al-Saeed Hospital. Conversely, there were 19 sterile samples or those that did not exhibit any bacterial growth. These samples included 5 from the private hospital and 14 from the government hospital, as seen in Fig. 2.

Out of all the samples, 83 (57.24%) of *Staphylococcus aureus* bacterial type were isolated. Among these, 51 (58.62%) originated from Misrata Medical Center (public hospital), and 32 (55.17%) from Al-Saeed Private Hospital. Additionally, 15 (17.2%) *Bacillus spp* samples were obtained from a public hospital, 19 (32.75%) from a private hospital, with 34 (23.44%) isolates originating from both hospitals. *Klebsiella spp* was isolated in 11 samples (7.59%) of the total, with 10 (11.49%) from the general hospital and one isolate (1.73%) from a private hospital. Furthermore, five samples (3.45%) of *Pseudomonas spp* bacteria were isolated, including four samples (4.60%) from the public hospital and one sample (1.73%) from the private hospital. *Streptococcus pneumonia* was present in six samples (4.13%), with two samples (3.45%) originating from the private hospital and four samples (4.60%) from the public hospital. Additionally, three isolates (2.07%) of *Staphylococcus epidermidis* were identified, all from the public hospital, with none found in the private hospital. *Escherichia coli* bacteria were exclusively found in the private hospital, totaling 3 samples (2.07%) of the overall samples, representing 5.17% of samples from the private hospital, as indicated in Table 2 and Fig. 3.

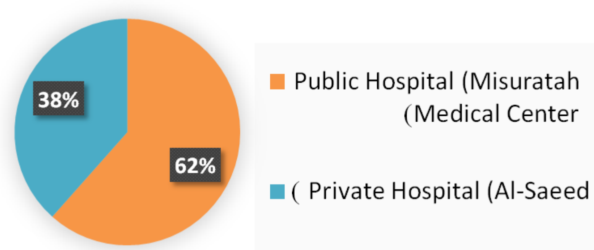


Fig. 1: illustrates the distribution of samples collected from both hospitals

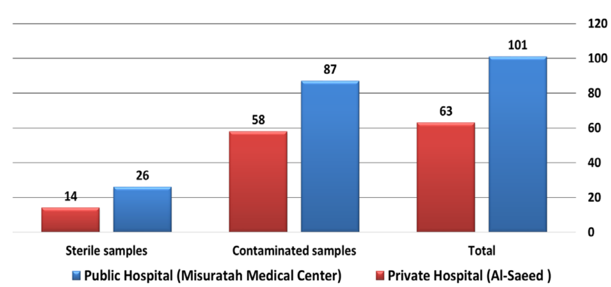
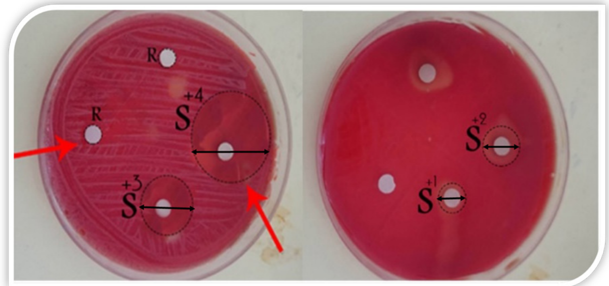


Fig. 2: presents the total number of samples, along with the count of contaminated and sterile samples, across both facilities

Table 2: Number and types of isolated bacterial strains and their percentage in both facilities

No	Bacteria	Number of isolates	Percentage
1	*Staphylococcus aureus*	83	57.24%
2	*Bacillus spp*	34	23.45%
3	*Klebsiella spp*	11	7.59%
4	*Streptococcus pneumonia*	6	4.13%
5	*Pseudomonas spp*	5	3.45%
6	*Staphylococcus epidermidis*	3	2.07%
7	*Escherichia coli*	3	2.07%
	Total	145	100%



S^{+4} = More than 20mmm, S^{+3} = From 15mm to 20mm, S^{+2} = From 10mm to 15mm, S^{+1} = From 8mm to 10mm, R = Resistant

Fig. 4: Shows the gradation of sensitivity to disinfectants against bacterial species

In testing the sensitivity of disinfectants and sterilizers on isolated bacterial species, it was observed that many of these bacteria exhibited sensitivity to most of the sterilizers and disinfectants used, although resistance to some emerged. The sensitivity gradation of these bacterial species was categorized based on the diameter of the zone of inhibition surrounding the filter paper disc for these materials, measured in millimeters. The highest sensitivity level, denoted as S^{+4}, corresponds to a zone of 20 mm and above, followed by S^{+3} for zones measuring between 15 and 20 mm, and S^{+2} for zones between 10 and 15 mm. The least sensitive category, labeled as S^{+1}, encompasses zones ranging from 8 to 10 mm, indicative of weak sensitivity. Any absence of inhibition zones is classified as resistance (R), signifying that the material has no effect on the bacteria (Fig. 4).

Table 3 presents an overview of the effects observed with some of the sterilizers and cleaners utilized, including commercial Clorox and Dettol, Ethanol, Propanol AF, Popular sodium hypochlorite (PSH), and Minuson AF. It also explores the potential interaction between the two sterilizers when combined, examining whether the effect is inverse or synergistic. This investigation stems from findings indicating that the two hospitals initially used popular sodium hypochlorite, followed immediately by one of the other sterilizers, without allowing for a waiting period, leading to the mixture of the two compounds. It was observed that the effectiveness of each compound varied across different types of bacteria, with sensitivity levels ranging from highly sensitive (S^{+4}, S^{+3}), moderate (S^{+2}), weak (S^{+1}), to resistant (R). Specifically, *Staphylococcus aureus* demonstrated high sensitivity to popular sodium hypochlorite, Minuson AF, and Ethanol. When Popular sodium hypochlorite (PSH) was mixed with Minuson AF or

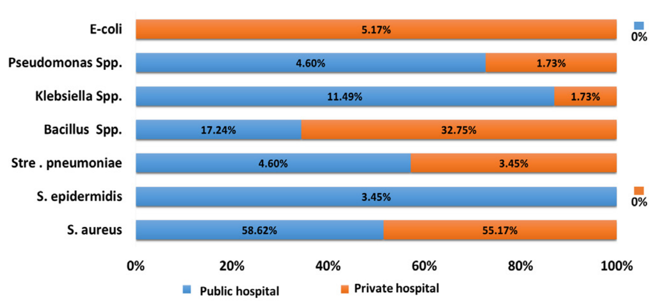


Fig. 3: illustrates the percentage of isolated bacteria for each type in both hospitals

propanol AF, or when Minuson AF was mixed with Ethanol, Clorox, and Dettol, it exhibited high sensitivity. However, it showed weak sensitivity to propanol AF mixed with Ethanol and was unaffected by propanol AF alone. On the other hand, *Staphylococcus epidermidis* bacteria were resistant to Popular sodium hypochlorite (PSH) and propanol AF but highly sensitive to Minuson AF alone and Ethanol. They also showed high sensitivity when Minuson AF was mixed with popular sodium hypochlorite, Ethanol, Clorox, and Dettol. *Streptococcus pneumonia* bacteria exhibited resistance to Propanol AF, Propanol AF mixed with Ethanol, and weak sensitivity to ethanol without mixing, Minuson AF mixed with popular sodium hypochlorite, and Dettol. They demonstrated high sensitivity to popular sodium hypochlorite, Minuson AF alone, Propanol AF mixed with Minuson AF, Minuson AF mixed with Ethanol, and Clorox. Interestingly, there was no improvement observed when Minuson AF was mixed with Propanol AF or with Ethanol. On the other hand, *Bacillus spp* showed sensitivity to Ethanol and Minuson AF mixed with popular sodium hypochlorite, and less sensitivity to Popular sodium hypochlorite (PSH) alone, as well as to Propanol AF and Minuson AF. They also exhibited sensitivity to Propanol AF mixed with Ethanol, Minuson AF mixed with Ethanol, and Clorox. Additionally, they showed weak sensitivity to commercial Dettol and weak resistance to Propanol AF mixed with Minuson AF. *Klebsiella spp*. exhibited high sensitivity to commercial Clorox, Minuson AF, Minuson AF mixed with Propanol AF, and Minuson AF mixed with Ethanol. They also showed moderate sensitivity to Minuson AF mixed with Popular sodium hypochlorite (PSH) and commercial Dettol. However, they were resistant to popular sodium hypochlorite, Propanol AF, Ethanol alone, and Ethanol mixed with Propanol AF. Interestingly, an opposite effect was observed when Popular sodium hypochlorite (PSH) was mixed with Minuson AF against this bacteria. Popular sodium hypochlorite, commercial Clorox, and Ethanol each exhibited a high effectiveness against *Pseudomonas spp* bacteria. Additionally, Propanol AF, Minuson AF, Propanol AF mixed with Ethanol, Minuson AF mixed with Ethanol, and Propanol AF mixed with Minuson AF showed a moderate effect on these bacteria. However, there was no observed effect when Popular sodium hypochlorite (PSH) was mixed with Minuson AF against *Pseudomonas spp*.*E. coli* bacteria demonstrated resistance to Propanol AF alone and Propanol AF mixed with Ethanol. Conversely, they exhibited high sensitivity to Minuson AF, Minuson AF mixed with popular sodium hypochlorite, Minuson AF mixed with Propanol AF,

Table 3: presents the efficacy of disinfectants and combined compounds against the isolated bacteria

Bacteria types	*Sterilizers and disinfectants used*				*Alternative disinfectants and sterilizers*		*Combined disinfectants and sterilizers*			
	PSH	*Propanol*	*Minuson*	*Ethanol*	*Clorox*	*Dettol*	*Minuson + Ethanol*	*Propanol + Ethanol*	*Propanol + Minuson*	*PSH + Minuson*
Staphylococcus aureus	S^{+3}	R	S^{+4}	S^{+3}	S^{+4}	S^{+3}	S^{+4}	S^{+1}	S^{+4}	S^{+3}
Bacillus spp	S^{+2}	S^{+3}	S^{+3}	S^{+4}	S^{+3}	S^{+1}	S^{+3}	S^{+2}	R	S^{+4}
Klebsiella spp	R	R	S^{+3}	R	S^{+4}	S^{+2}	S^{+3}	R	S^{+3}	S^{+2}
Streptococcus pneumonia	S^{+4}	R	S^{+3}	S^{+1}	S^{+3}	S^{+1}	S^{+3}	R	S^{+3}	S^{+1}
Pseudomonas spp	S^{+4}	S^{+2}	S^{+2}	S^{+3}	S^{+4}	S^{+1}	S^{+2}	S^{+2}	S^{+1}	R
Staphylococcus epidermidis	R	R	S^{+4}	S^{+3}	S^{+4}	S^{+4}	S^{+3}	S^{+1}	S^{+2}	S^{+4}
Escherichia coli	S^{+2}	R	S^{+4}	S^{+2}	S^{+3}	S^{+3}	S^{+4}	R	S^{+4}	S^{+3}

Minuson AF mixed with Ethanol, and commercial Dettol. Moreover, they showed moderate sensitivity to Popular sodium hypochlorite (PSH) and Ethanol when used separately (Table 3).

DISCUSSION

In the current study, we observed the presence of pathogenic bacterial species in newborn incubators (*Staphylococcus aureus*, *Bacillus* spp, *Klebsiella* spp, *Pseudomonas* spp, *Streptococcus pneumonia, Staphylococcus epidermidis, and Escherichia coli).* The isolated species was consistent with the findings of multiple studies, including [4, 6, 11, 12, 19], which confirmed the presence of the same bacterial species with a few minor variations in one or two types of bacteria. The presence of such species is regarded as a significant hazard in the field of infection control, as in a study conducted by Bhatta *et al.*'s that stressed the risk of bacterial contamination in newborn incubators [11]. Particularly because it is in incubators for newborns with a variety of disorders, including physical deficiencies, irregular heartbeats, immune deficiencies, etc., which makes the issue more difficult to solve and leads to a decline in health [9, 10]. Our study's findings align with the research conducted by Asinobi *et al.*, who reported that *S. aureus*, a multidrug-resistant bacteria, was the most frequently isolated species from newborn incubators in intensive care units [2, 12]. It's noteworthy that the bacterial species isolated in our study closely resemble those found in other studies conducted in various locations, including incubators and neonatal intensive care units. These results are in line with the species that Bokulich *et al.* isolated from neonatal blood samples[10]. This provides proof of hospital-acquired infections and emphasizes the inadequacy of sterilization procedures, which are crucial in determining morbidity, mortality, and the subsequent healthcare costs in these environments.

In the assessment of disinfectant efficacy against the isolated bacterial strains, Minuson AF demonstrated effectiveness against all strains, suggesting its suitability for sterilization and infection control measures. Popular sodium hypochlorite (PSH)was effective against all species except *S. epidermidis* and *Klebsiella spp*. Ethanol exhibited effectiveness against all isolated bacterial species except *Klebsiella spp*, with weaker efficacy noted against *Streptococcus pneumoniae*, this is likely consistent with the findings of Rasool and Ibrahim, wherein ethanol was observed to decrease bacterial species isolated from neonatal intensive care unit (NICU) computers compared to before its application [22]. Conversely, Propanol AF displayed ineffectiveness against all isolated species except *Bacillus* spp and *Pseudomonas* spp, warranting a reevaluation of its usage, which is partially aligned with the study conducted by Elrotob *et al.*, who investigated the effectiveness of this disinfectant against *Pseudomonas* spp [21].

In an effort to enhance infection control, Minuson AF disinfectant was tested in combination with other disinfectants and sterilizers to assess potential synergies or drawbacks. When combined with Popular Sodium Hypochlorite, the latter's effectiveness improved against various bacterial species such as *Bacillus* spp*, S. epidermidis*, *E. coli*, and *Klebsiella* spp. However, this combination compromised the effectiveness of Minuson AF against all isolated species except *Bacillus* spp. Consequently, it is advised against using these two compounds together or consecutively, as Popular sodium hypochlorite (PSH) negatively impacts the efficacy of Minuson AF. Combining Minuson AF with Propanol AF resulted in an enhancement of Propanol AF's effectiveness against all isolated strains except *Pseudomonas* spp and *Bacillus* spp, in comparison to Propanol AF used alone. However, this combination had a negative impact on the effectiveness of Minuson AF against strains such as *S. epidermidis*, *Bacillus* spp, and *Pseudomonas* spp, when compared to the efficacy of Minuson AF alone. Consequently, due to these conflicting effects, this combination appears impractical for infection control purposes. Combining Propanol AF with Ethanol resulted in a slight enhancement of Propanol AF's effectiveness against *S. aureus* and *S. epidermidis*, albeit with decreased effectiveness against *Bacillus* spp compared to Propanol AF used alone. However, this combination decreased the effectiveness of Ethanol against all isolated strains, rendering it an improbable choice for infection control purposes. When Minuson AF was combined with Ethanol, an enhancement was observed in ethanol's effectiveness against these isolated strains. Moreover, this combination did not alter the efficacy of Minuson AF against these species, thereby maintaining its efficiency. Hence, this combination can be utilized as Minuson AF complements the effect of Ethanol.Given the absence of previous studies on combining these disinfectants and sterilizers, these results provide crucial evidence regarding the positive or negative effectiveness of such combinations. This holds particular significance, especially in scenarios where these materials are randomly utilized or in facilities where detergents like sodium hypochlorite are used first, potentially followed by another sterilizer or disinfectant, leading to suboptimal sterilization outcomes. Conversely, such combinations might enhance sterilization, particularly in the face of increasing bacterial resistance. However, this necessitates further in-depth studies focusing on the mechanisms and specifics of these combinations. On the other hand, the outcomes showed that Clorox and Dettol were effective against every isolated strain, suggesting that they could be used as substitute materials during the sterilizing process. These findings are in line with study by Saha *et al.* that demonstrated the potency of Dettol against isolated microorganisms [23], and by Ozturk *et al.* that demonstrated the potency of Clorox (sodium hypochlorite 10%) against bacteria that were isolated [24].

The variability in effectiveness and sometimes the ineffectiveness of these disinfectants and sterilizers may stem from various factors, including product quality, sourcing, dilution levels, and storage conditions. This observation has been supported by several studies. For instance, Aminu and Abdulhadi highlighted the importance of adhering to proper dilutions of sterilizers and disinfectants to ensure their efficacy in pollution and infection control [25]. Elrotob *et al.* underscored the significance of considering product quality regarding the storage and utilization of these compounds, advocating against the continual use of the same compound and concentration to prevent bacterial resistance[21]. Furthermore, Prisce *et al.* emphasized the correct utilization of disinfectants and sterilizers according to the accompanying information, stressing the importance of continuous monitoring, especially regarding storage conditions. Their study demonstrated that freshly prepared disinfectants and sterilizers exhibit greater effectiveness compared to those that are stored [4]. This underscores the necessity for ongoing vigilance and monitoring of the quality of disinfectants and sterilizers to ensure the efficacy of sterilization procedures.

CONCLUSION

Hospital-acquired infections pose a significant global challenge, leading to the proliferation of severe diseases and elevated mortality rates. Newborns, with their developing immune systems and potential accompanying vulnerabilities such as low weight or blood pressure, are particularly susceptible. The cleanliness and sterility of incubators and their surroundings are crucial to safeguarding these vulnerable infants. Results of our study conducted in two major hospitals in Misurata revealed the presence of various pathogenic bacteria in newborn incubators and deficiencies in the efficacy of certain disinfectants and sterilizers. The findings also indicated that Minuson AF, Clorox, the combination of Minuson AF and Ethanol, and Dettol were the best disinfectants and sterilizers that had an impact on these isolated species. Additional research and analysis are required to determine the efficacy of the remaining disinfectants and sterilizers with regard to usage, storage, handling, and source quality. These findings underscore the urgent need to address hospital-acquired infections, improve sterilization protocols, and periodically assess the effectiveness of disinfectants used in healthcare settings.

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CONSENT FOR PUBLICATIONS

The authors have reviewed the manuscript and grant permission for the publication of its final version.

CONFLICT OF INTEREST DISCLOSURE

The authors confirm that they have no conflicts of interest to disclose.

AVAILABILITY OF DATA AND MATERIAL

The authors can provide data upon request.

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