A Study about Bacterial Contamination In Maternity and Children Teaching Hospital.

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الخلاصة

أظهرت نتائج دراستنا أن نسبة التلوث البكتيري في العمليات قبل عملية التطهير والتعقيم كانت (66.2 %) وبعد عملية التعقيم انخفضت إلى (47.5 %) ، وإن أعلى نسبة للتلوث كانت في غطاء الأرضيات (100 %) وانخفضت إلى (60 %) بعد عملية التعقيم والتطهير، وإن أقل نسبة للتلوث كانت في الأدوات الجراحية (1%) قبل عملية التعقيم ولم يظهر أي تلوث في هذه الأدوات بعد عملية التعقيم وفيما يخص المطهرات والمعقمات فكانت نسبة التلوث فيها (55 %) وأعلى نسبة للتلوث (75 %) كانت في مطهر الديتول، وأقل نسبة (25 %) كانت في الفورمالين.

Abstract

The results of our study showed that percentage of bacterial contamination in operating rooms was (66.2%) before disinfection method, and after disinfection method was (47.5%). High contamination percentage (100%) was in room floor covers and was decreased to (60%) after disinfectant. The lowest percentage of contamination (1%) was in surgical instruments and disappeared after disinfectant. The percentage of contamination in disinfectants and antiseptics was (55%), high contamination percentage (75%) was in Chloroxylenol (Dettol) whereas the lowest (25%) was in Formalin.

Introduction

After AIDs and food borne illnesses, nosocomial infections are the most costly and deadly infectious diseases, and are growing more dangerous each year. As more new pathogens emerge with resistance to available antibiotics, the rate of NIs in use has risen (36%) in the last two decades, bringing more than (40.000) deaths annually along with high costs in the treatment. There were about 20 million NIs in 2004, each adding one or more extra days in the hospital. A wound infection or surgical site infection, for example, adds about a week to the hospital stay, even though the average of NIs is relatively easy to death with, about (4%) of the patients will die. So, if it is possible to cut out 100 of those NIs in the hospital, it is possible also to cut out four patient deaths (13).

The factor that contributes to the risk of acquiring an NI includes severity of the illness frequency of invasive diagnostic and therapeutic procedure variation in the effectiveness of infection control programs. The poor state of health of many patients, the use of immuno-supportive therapy, extensive surgery invasive diagnostic tests, use of indwelling catheters in veins, arteries and bladder, indwelling tubes in the respiratory therapy equipment, and the wide spread use of extended-spectrum antibiotics, which leads to the prevalence of antibiotics resistant-strains of bacteria in the hospital environment (6)

Other factors such a poor nutritional status, extensive burns, wound and young and elderly are generally more susceptible to infection.(10).

For these reasons, this study is designed to fulfil the following goals:

1. To isolate and define aerobic bacteria from operating rooms, kitchen environment.

2. To test the efficiency of disinfectants and antiseptics in-use.

Methods

1) Sterilization Procedure

Ordinary culture media and glass wares were sterilized by autoclaving at 121℃ (15 1b/inch) for 15-20 minutes, compounds or solutions sensitive to heat such as sugars, and urea solutions were sterilized by adding a little amount of chloroform to them and were shaken well then left to stagnate for 1 hr. thus these solutions became sterile. (12).

2) Sample Collection

The samples shown in table 1 were collected from 2 different sources in Delivery and Children Teaching Hospital in Al-Diwaniya Province from July 2003 to July 2004, these samples were collected at intervals before and after chemical disinfection by taking swabs according to the method that was suggested by (2).Sterile cotton swabs were soaked in sterial normal slaine solution (0.85%) and rolled in the test place and were carried to the laboratory of Medicine College for bacteriological testing.

A/ Operating Rooms Environment

The samples shown in table (2) were collected from operating rooms before and after chemical disinfection

Table (1): Sources and Numbers of all Tested Samples

Rank	Sources	No. of Tested Sample
1	Anesthesia mask	10
2	Rooms, floors and walls	20
3	Recovery instrument	10
4	Surgical instrument	10

5	Assistant cloths	10
6	Surgical operating beds	20
Total	6	80

Table (2): Numbers and Places of Samples Collected from Operating Rooms

B/ In-Use Disinfectants and Antiseptics

One ml. of each disinfectant or antiseptic was collected in sterile bottle and immediately taken to the bacteriology laboratory (table 3), and then this sample (one ml. from concentrated or random diluted disinfectant or antiseptic) was added to 9ml. diluent (Brain-hart infusion broth) and then incubated under aerobic condition at 37℃ for 48 hrs., then subcultured by using a sterile Pasteur pipette. 10 drops of each disinfectant or antiseptic/dilutent (mixture) were separately dropped into the surface of each of the following plates: nutrient agar, blood agar, MacConkey agar, EMB agar, Mannitol salt agar, and chocolate agar plates. These plates were incubated under aerobic condition at 37℃ for 3 days.(5)mentioned that the growth of more than 5 colonies/plate (i.e., 250 live bacteria/ml) on either of the plates means a failure in disinfection process or contamination of disinfectants or antiseptics from their places or sources.

Table (3): Types and Numbers of Samples from disinfectants and Antiseptics In-Use included in this Study

Rank	Disinfectants and Antiseptics	No. of Tested Sample
1	Spirit	20
2	Chloroxy Lenol	20
3	Chlorhexidine-gluconate	20
4	Formalin	20
Total	4	80

3) Diagnosis

All collected samples were cultured only aerobically and bacterial isolates were diagnostic by depending on morphological characters, which include shape of colony, edge of colony, fermention of Lactose sugar and nature of staining by Gram's stain. In addition to biochemical testes by using IMVIC test and fermention of sugar and API system.(3).

Results and Discussion

1) Operation Rooms Environment

Samples from different locations in operating rooms were collected before and after chemical disinfection (table 4). The contamination rate in operating room was (66.2%) before disinfection and after that became (47.5%). The contamination rate in recovery rooms floors before disinfection was relatively high (100%) in comparison with contamination rate after disinfection (60%), and the contamination rate in surgical operating beds accounted (85%) before disinfection, while it was reduced to (70%) after disinfection. Similar contamination frequencies were found in room floors and walls, anesthesia mask, assistant cloths and surgical instrument, which does not appear to have any contamination in it after disinfection. The most frequent isolates before disinfection among rooms floors and

walls, recovery room, surgical operating beds, anesthesia mask, assistant cloths were (13) *E.coli*, (13), *Proteus sp.*, (10) *CNS*, (8) *Staphylococcus sp.*, (5) *Klebsiella*, (3) *Citrobacter*, and (2) *Pseudomonas sp.* Surprisingly, the bacterial species isolated from samples after disinfection were different, this could be attributed to the carelessness through preparation of chemical disinfection (Formalin was used in approximate dilutions by tap water). It is clear that tap water contains *Enterobacter sp.*, *Providenciae*, *Pseudomonas*, *Serratia sp.*, *Acintobacter sp.* And other organisms. (1). These bacteria might resist the disinfectant or even multiply in those disinfectants causing contamination of it. (9) mentioned that contamination of tap water used for surgical hand washing is common. For checking up the sterility of surgical instruments were examined as shown in table (4) only one isolate of E.coli was found in among instruments. These materials should be extremely sterilized by autoclaving (11). The detection of E.coli in this material may indicate in efficient autoclaving due to the contamination of this material after sterilization (9)

Airborne bacteria transported via instruments is a more important cause of wound contamination than direct fall-out from the air (4). However, the contamination of operating environment might be ascribed to: dilution of disinfectant, its quality and activity, the storage conditions of disinfectant, regular timetable for disinfection, carelessness of hygienic rules, and absence of surveillance cadre in those rooms. Moreover, dirty debris had been seen on operating beds in the emergency operating rooms.

Table (4): Percentage of Bacterial Contamination Detected in Operating Rooms Beds

	Before Disinfection				After Disinfection			
Location	No. of Samples	No. of Contam.	%	No. of Bacteria Isolates	No. of Samples	No. of Contam.	%	No. of Bacteria Isolates
Room floors & walls	20	15	75	(3) *Proteus mirabilis* (3) *CNS* (3) *Citrobacter freundii* (2) *Klebsiella pneumoniae* (1) *Proteus vulgaris*	20	10	50	(3) *K. oxytoca* (1) *Staph. aureus* (1) *E.coli* (1) *Enterobacter cloacae* (2) *Klebsiella aerogenes*
Recovery rooms floors	10	10	100	(5) *CNS* (3) *E.coli* (2) *Ps. fluorescens*	10	6	60	(2) *Ps. aeruginosa* (2) *E.coli* (1) *providencia sp.*
Surgical operating beds	20	17	85	(5) *Staphylococcus aureus* (4) *Proteus mirabilis* (3) *Klebsiella pneumonia* (3) *E.coli*	20	14	70	(3) *Serratia marcescens* (3) *Proteus penneri* (3) *Kl. oxytoca*
Anesthesia mask	10	4	40	(2) *E.coli* (2) *CNS*	10	3	30	(2) *Providencias sp.* (1) *Ps. aeruginosa*
Assistant cloths	10	3	30	(2) *Proteus vulgaris* (1) *E.coli*	10	2	20	(1) *CNS* (1) *Ps. aeruginosa*
Surgical instrument	10	1	10	(1) *E.coli*	10	0	0	
Total	80	53	66.2	51	80	38	47.5	36

2) Sample Taken from Disinfectants and Antiseptics In-Use

In attempt to detect the bacterial contamination in each disinfectant or antiseptic used in the hospital, some of concentrated (undiluted)and diluted samples from each disinfectant or antiseptic were tested as shown in tables (6, 7, and 8) for spirit, chloroxylenol, chlorhexidine-gluconate, and formalin.

Of (80) disinfectants and antiseptics in-use (44) (55%) were contaminated as shown in table (5). A high level of contamination in random diluted disinfectants and antiseptics has been found relatively high contamination rate (75%) was found in chloroxylenol (Dettol), while in formalin was (25%). We noticed during this study that all diluted disinfectants and antiseptics were kept and used for several weeks in the wards, rooms and laboratory. The results varied according to various factors, such as the life of disinfectants deteriorate when diluted with water (3). In correct measurement of disinfectant or of water when making up the dilution, keeping the dilution in use for several days, refilling empty containers without cleaning and drying them, "topping up" a container in which the dilution has been partly used, repeated use of the same container of disinfectant over an extended period of time, the presence of an inactivating material prolonged storage, storage conditions, the choice of disinfectant, which is not good one for the particular purpose, and bacteria from the caps and the area around the opening of container may contaminate the disinfectants it holds (5).

All these factors lead to contaminate disinfectants and antiseptics. Several samples of original concentrated of disinfectants and antiseptics were found contaminated in this study, this may be due to; contamination of raw material used in the structure of disinfectant, contamination of the material exploited in the stages of industrialization, air, contamination of the operator hands, and jars used for filling. Both(7) noticed that CHX used in the hospitals had lost its validity and it was still in the primary jar

stage, that was caused by existence of natural inhibitory materials in the corks of jars.

Table (5): Percentage of Contaminated Disinfectants and Antiseptics

Disinfectants & Antiseptics	No. of Samples Tested	No. of Contaminated	%
Spirit	20	11	55
Chloroxylenol	0	15	75
Chlorhexidine-gluconate	20	13	65
Formalin	20	5	25
Total	80	44	55

In our study, strains of Ps-aeruginosa were found in each spirit, chloroxylenol, chlorhexidine-gluconate and formalin. This microorganism was described as a strong resistant to various disinfectants, antiseptics, and other antimicrobial agents, for this it is referred to as a multi-drug resistant microorganism (8).

The results of the current study indicated that Staphylococcus sp. was found in all disinfectants and antiseptics, which tested in our study. Disinfectants and antiseptics resistance is encoded in multi-resistant plasmid determinants of staphylococcus. Extensive use of disinfectants and antiseptics has been implicated in the evolution and persistence of the multi-resistant chromosome of staphylococcus sp. And its continued use could encourage persistence and spread of combined antiseptic-antibiotic multiple resistance in *Staphylococcus sp.* (11). Gram-negative bacteria was also found in disinfectants and antiseptics. These bacteria include; *E.coli*; *Proteus sp.*; *Providanciae sp.*; *Serratia sp.*; *Citrobacter sp.*; *Enterobacter sp.* and *Acintobacter sp.* These results may confirm the results reported by (6).Resistance of these strains to disinfectants and antiseptics may be due to the subtle changes in the structural arrangement of the cell envelopes of these strains which was associated with this resistance and the inner membrane was not implicated (8).

Table (6): Contamination of In-Use Spirit by Bacteria

Source	No. of tested	No. of randomly diluted samples	No. of contaminated %	Bacterial contaminants	No. of concentrated Samples	No. of Contaminated %	Bacteria contaminants
Operating rooms	8	6	2(33.3)	*(2) Staph. aureus*	2	1(50)	*(1) CNS*
Emergency ward	6	6	3(50)	*(1) Ps. aeruginosa* *(1) Citrobacter sp.* *(2) CNS*	-	-	
Patient wards	6	6	5(83.3)	*(2) Ps. aeruginosa* *(2)Acintobacter calcoaceticus* *(2) Providanciae sp.*	-	-	
Total	20	18	10(50)	12	2	1(10)	1

Table (8): Contamination of In-Use Chlorhexidine-Gluconate by Bacteria

Source	No. of tested	No. of randomly diluted samples	No. of contaminated %	Bacterial contaminants	No. of concentrated Samples	No. of Contaminated %	Bacteria contaminants
Operating rooms	5	1	1(100)	*(1) CNS*	4	2(50)	*(1) Ps. aeruginosa* *(1) CNS*
Emergency ward	5	5	3(60)	*(1) E.coli* *(1) Providanciae sp.* *(1) CNS* *(1) Proteus mirabilis*	-	-	
Laboratory	5	5	3(60)	*(2) Ps. aeruginosa* *(1) Serratia sp.*	-	-	
Patient wards	5	4	4(100)	*(2) CNS* *(1) Staph. aureus* *(1) E.coli*	1	1(100)	*(1) E.coli* *(1) Enterobacter sp.*
Total	20	15	11(55)	12	5	2(10)	4

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