

Microbiological Assessment of Surgical Operations Rooms' Clothes

Yehia Abd El Moneim Osman

Prof. of microbiology and molecular biology, Botany Dept. Faculty of Science, Mansoura University, Egypt.

Zeinab Ahmed Abd El Aziz

Associate Prof. of Clothes and Textile, Home Economics Dept. Faculty of Specific Education, Mansoura University, Egypt.

Amal Fawzy Abd El Monem El Tantawy

Assistant Lecturer Home Economics Dept. Faculty of Specific Education, Mansoura University, Egypt.

Abstract:

The study problem identified in dealing with the subject of an important type of functional clothing, that are used in an important and significant sector such as the medical sector in Egypt, with no knowledge of the health damage that may be caused by the use of such clothing as a result of erroneous transactions that take place upon due to lack of possibilities, since the devices washing and sterilization are often available in specialized centers without the other, so the medical clothing pass is codified practices.

Due to the low level of transactions conducted on the medical clothing in many hospitals, and non-compliance with specifications selected, it should pay attention to the need to upgrade these transactions, and to develop mechanisms that will raise the level of health awareness among medical clothing users. The study attempted to answer following questions (1) What methods used to deal with medical clothing used in operating rooms in hospitals? (2) What damages that can result from the use of medical clothing used in operating rooms? (3) Which is better healthier medical clothing manufacturer of woven fabrics or manufacturer of non-woven fabric? The study aimed at evaluating methods of dealing with medical operating rooms' clothes in some hospitals (sterilization and disinfection), due to determine whether medical operating rooms' clothes cause health damage to its users or to others or not, also evaluating of medical clothing manufacturer of woven fabrics, and manufactured of non-woven fabrics, which used in the operating rooms of some hospitals and finally identify the vision of specialists in the field of medical field in the level of medical clothing, used in operating rooms in some hospitals.

Keywords:

Surgical Gowns
Medical Clothing
Operating Rooms' Clothes
Infection

Paper received 25th April 2017, Accepted 13th May 2017, Published 1st of July 2017

1- Introduction:

Clothes are a source to protect the human and from the various factors that are harmful to his health, they are also considered the most important reasons that may bring a lot of health damage and numerous diseases, as one of the most important functions' clothes is to cover the body human to allow its protection from all harmful by the external influence of environmental factors and variables.

Clothes' products at the present time enjoy intensive careful from researchers and practitioners in the field of development and improvement of fabrics produced them these clothes, so as to meet the requirements of use and human consumption of fabrics, although the industrial enormous progress that accompanied the Industrial Revolution led to the emergence of new varieties of raw materials and chemicals to improve the properties of the qualities and requirements needed by the consumer, but it is

possible to isolate a huge number of proliferating microorganisms in the clothes fitted and treatment of chemical, mechanical and materials that have been shown to have damaged undesirable impact on health, as they adversely affect the properties of the fabric.⁽⁸⁾

Medical textiles sector is one of the most successful areas in recent times, so this fabrics and which related health care products is limited to bandages and ligaments, especially made of cotton and viscose fabrics used in hospitals, for example, patients' clothes and bedding and diapers, but lately both market size and diversity of the products available has increased.⁽⁵⁾

The fabrics used in the medical fields are considered types of fabrics that we have been interest in them to catch up with advances in technology and textile design, which dictates the need to develop and raise the efficiency of the performance of these medical fabrics to bring it to quality check its global competitiveness level, and

product quality is determined by the extent appropriate and fit the actual characteristics of the fabric to the requirements of use and convenient functionality that produced it, functional appropriate is determined by the study of nature and conditions of employment, and medical textile products must have quality standards provide safety for patients and medical personnel, and medical textile products and its' functional performance are evaluated according to specific conditions and specifications.⁽⁴⁾

C.L. Shelton, *et al.*, (2010) entitled said that healthcare workers' clothes can become contaminated with pathogens. This study aimed to determine whether hospital doctors can change their attire to reduce the possibility of cross-infection without jeopardising the doctor-patient relationship. In this experimental repeated-measures study, 55 male and 45 female inpatients (mean age: 52.89 years) were randomly selected. Participants were shown photographs of medical dresscodes and asked to rate their appropriateness on a 5-point Likert scale. They were then read a standardised statement regarding the significance of attire to cross-infection and the exercise was repeated. Pre statement, there was no significant difference between the majority of dress options, though casual dress (male and female) and short-sleeved top (female) were considered significantly less appropriate. Post statement, surgical 'scrubs' and short-sleeved top/shirt were deemed most appropriate ($P < 0.0001$). There was no significant difference between short-sleeved shirt and scrubs for males. For females, scrubs were preferred ($P = 0.0005$). Participants expressed no significant preference for medical attire, with the exception of a dislike of casual dress. However, once informed of the significance of attire to cross-infection, preference changes to favour dresscodes which may minimise infection risk.⁽²⁾

Finally Yonit Wiener-Well, *et al.*, (2011) reached that a total of 238 samples were collected from 135 personnel, including 75 nurses (55%) and 60 physicians (45%). Of these, 79 (58%) claimed to change their uniform every day, and 104 (77%) defined the level of hygiene of their attire as fair to excellent. Potentially pathogenic bacteria were isolated from at least one site of the uniforms of 85 participants (63%) and were isolated from 119 samples (50%); 21 (14%) of the samples from nurses' gowns and 6 (6%) of the samples from physicians' gowns ($P = NS$) included of antibiotic-resistant bacteria.⁽¹¹⁾

2- Materials and methods:

This side includes laboratory tests and which has been selected study sample represented by a

sample of (20) a multi-use medical gown compiled from various stages of their use from Mansoura University in Dakahlia Governorate, an organism or more of each sample have been isolated, The total number of organisms isolated reached (35) organism, and these samples were tested to detect the presence of certain types of bacteria that can be found in the clothing examples of Staphylococcus Aureus, Gram negative bacteria, which can be considered one of the causes of skin diseases, which can be transmitted by clothes, then make a definition of these bacteria to identify their types, laboratory tests have been conducting in bacteriological laboratory at the Faculty of Specific Education, Mansoura University, definition tests were conducted at the Children's Hospital at Mansoura University, as well as some confirmatory tests were conducted at Faculty of Science at University Mansoura.

3- Materials

Sample (bacterial and plasmid):

This study was carried on bacteria isolated from a wastewater samples collected from Mansoura hospitals (Mansoura University Children's Hospital – Specialized Medical Hospital – Oncology Center - Emergency Hospital – Mansoura University Hospital) to check the sanitary of physician coats and these samples were numbered from (1) to (20), one organism or more was isolated from each sample, where the total number of isolated organisms from samples reached (35) of an organism.

Bacteriological medium:

All media were prepared in one liter distilled water and autoclaved at 121 °C for 20 minutes, and store at room temperature until use as recommended by Difco Laboratories.

Luria-Bertani broth (LB):

Tryptone	10.0 gm
Yeast extract	5.0 gm
Sodium chloride	10.0 gm
Distilled water	1 Liter
pH was Adjusted to 7	

LB Agar

LB- broth was solidified by adding 15 gm agar per liter of LB broth.

Staining reagents

1-Crystal violet solution

a- Solution A

Crystal violet	1.0	gm
Distilled water	10.0	ml

b- Solution B

NaHCO ₃	1.0	gm
Distilled water	10.0	ml

For use mix 1.5 ml of solution A with 0.5 ml of solution B.

2- Safranin solution

Safranin powder	0.25	gm
Distilled water	250.0	ml

Diluted 10-15 time its water.

Sodium dodecyl sulfate polyacrylamide gel (SDS-PAGE)

Polyacrylamide gel consisted of two layers: resolving or separating gel (lower layer) and stacking or sample concentrating (upper layer) in which comb or a well former is immersed; each layer gel was prepared as indicated below (Laemmli, 1970).⁽⁶⁾

a- Preparation of separating gel (12%)

Distilled water	3.0125	ml
1.5 M tris HCl	1.875	ml
10% SDS	75	µl
Acrylamide /bis (30%)	2.500	ml
10% ammonium per sulfate (APS)	37.5	µl
TEMED	2.5	µl

Volume required to completely fill gel sandwich may be adjusted depending on size of application.

b- Preparation of stacking gel

Distilled water	1.525	ml
0.5 M tris HCl	625	µl
10% SDS	25	µl
Acrylamide /bis (30%)	333	µl
10% ammonium persulfate (APS)	20	µl
TEMED	2.0	µl

All reagent of both separating and stacking gel layers, except TEMED and APS, were mixed de-aerated under vacuum for two minutes, then the polymerization catalysts (TEMED and APS) were added immediately prior to casting the gel.

Stock solutions

1- Monomer solution

Acrylamide	58.4	gm
Bis-acrylamide	1.6	gm
Distilled water	200	ml

Store at 4 °C in the dark

2- (4x) Tris-CI 1.5M (separating buffer)

Tris base	36.3	gm
Distilled water	200	ml
Adjusted pH=8.8		

3- (4x) Tris-CI 0.5M (stacking buffer)

Tris base	3	gm
Distilled water	50	ml
Adjusted Ph=8.8		

4- Sodium dodecyl sulfate (SDS) 10%

SDS	10	gm
Distilled water	100	ml

5- Ammonium per sulfate (APS) 10%

Ammonium per sulfate	0.1	gm
Distilled water	1	ml

6- Running gel overlay (0.375 M Tris-CI, pH 8.8, 0.1% SDS)

Tris	25.0 ml from solution (2)
SDS	1 ml from solution (4)
Distilled water	100 ml

7-(2X) treatment buffer (0.125M Tris-CI, pH 6.8, 4% SDS, 20% glycerol, 10% β-M.E.)

Distilled water	10 ml
Tris-CI	2.5 ml from soln (3)
Glycerol	2 ml
10% SDS	4.0 ml from soln(4)
β-Mercepto ethanol (M.E.)	1.0 ml

8-Tank buffer (0.025 M Tris, pH 8.3, 0.192 Mg lycine, 0.1 SDS)

Tris-base	12	gm
Glycine	57.6	gm
10% SDS	40 ml from soln.(4)	
Distilled water	4	litter

9-Stain stock (1% Coomassie Brilliant Blue R-250)

Coomassie Brilliant Blue(R-250)	2	gm
Distilled water	200	ml

Stirred and filtered before storage and use.

10-Staining solution (Coomassie Brilliant Blue R-250, 0.1% Methanol 50%, Glacial acetic acid 10%)

Coomassie Brilliant Blue(R-250)	62.5	ml from soln.(9)
Methanol	250	ml
Glacial acetic acid	50	ml

11- Destaining solution

Glacial acetic acid	500	ml
Methanol	100	ml
Distilled water	1	L

Bromophenol blue was used as tracking dye and mixed with the breakage (treatment) buffer.

Protein marker (MW)

Spectra™ Multi color Broad Range protein ladder supplied by (Koma Biotech, Korea). MW marker consisted of 8 polypeptides (10,15, 25, 55,70,100,130 and 250 kDa) and stored at -20 °C .

Bacterial Isolation:

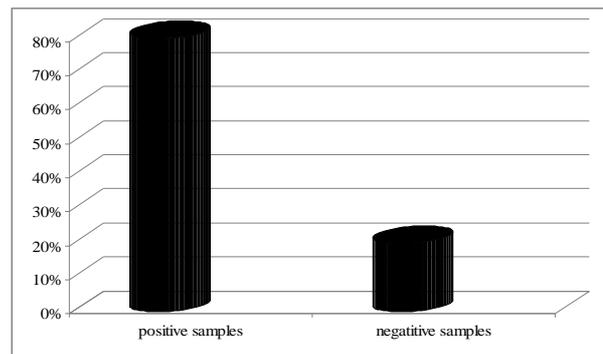
All samples were subjected to laboratory tests for the first phase to obtain pure bacterial colonies and definition according to Marisha Burden, *et al.*, (2011)⁽⁷⁾, the results of preliminary tests has confirmed emergence of bacteria in the number (16 samples) of the study samples at different

stages of use that have isolated them, and this as shown in figure (1).

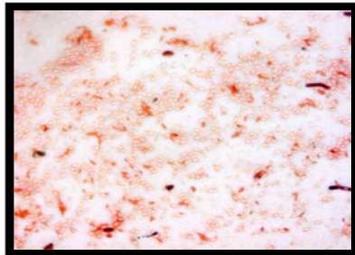
II) Methods

II) Methods : Microscopic examinations

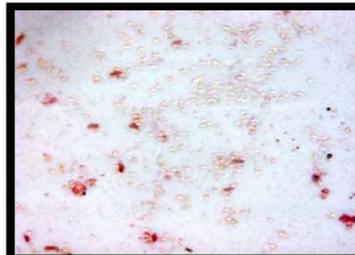
The purified bacterial culture was grown on LB agar medium and single colony was used to prepare a bacterial film for gram-staining. The stained bacterial film was examined for the Gram's stain susceptibility and shape of the cells (Benson *et al.*, 1998; Collee *et al.*, 1996; and Steinbach and Shetty, 2001).⁽³⁾



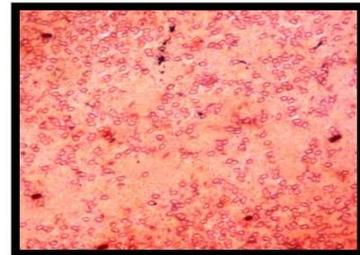
Figure(1) shows the presence of bacteria tested samples ratios



Picture (1) shows organism's shape (6-1- B) under microscope



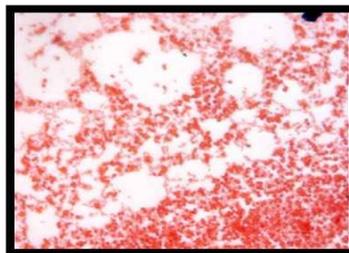
Picture (2) shows organism's shape (6-2) under microscope



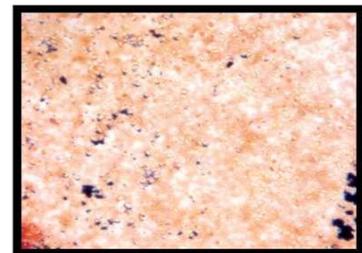
Picture (3) shows organism's shape (9-1) under microscope



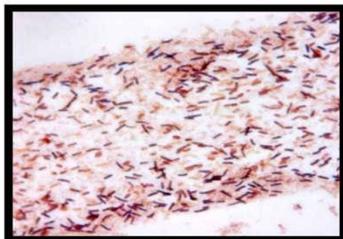
Picture (4) shows organism's shape (1-1) under microscope



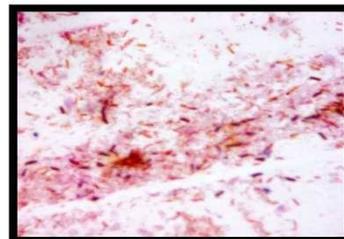
Picture (5) shows organism's shape (1-2) under microscope



Picture (6) shows organism's shape (6-1- A) under microscope



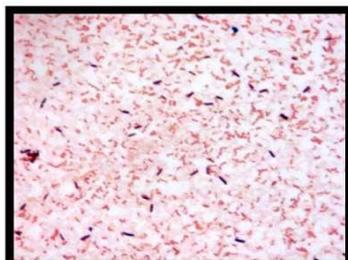
Picture (7) shows organism's shape (19-2) under microscope



Picture (8) shows organism's shape (18-1) under microscope



Picture (9) shows organism's shape (10-1) under microscope



Picture (10) shows organism's shape (19-5) under microscope



Picture (11) shows organism's shape (19-4- B) under microscope



Picture (12) shows organism's shape (19-4- A) under microscope

Bacterial Identification

I-Automated identification

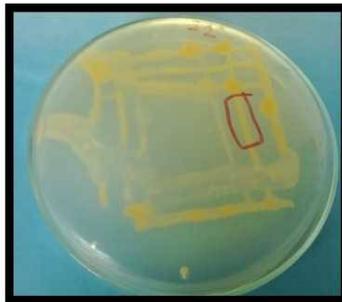
The identification of the bacterium in this study was confirmed by using MicroScan WalkAway - 90 Systems Dried Gram negative bacteria (trademark of Siemens Healthcare Diagnostics Ltd. Sir William Siemens Sq. Frimley, Camberley, UK GU16 8QD.). Pediatrics Hospital, Microbiology Lab., Mansoura University, Mansoura, Egypt which is an automated system incubates micro titer identification and antimicrobial susceptibility testing panels, interprets biochemical results through the use of a photometric or fluorogenic reader, and generates computerized reports that can be interfaced with hospital mainframe information systems. Conventional panels utilize the photometric reader and provide identification results for gram-negative bacilli within 15 to 42 h, with reagents

added automatically by the WalkAway instrument. Panels can be removed from the WalkAway instrument and read manually if verification is necessary. Panels for identification of gram-negative bacilli contain 26 modified conventional biochemical and six antibiotics. The database associated with the MicroScanWalkAway instrument contains information for the identification of 59 groups, genera, or species of members of the family *Enterobacteriaceae* and 57 groups, genera, or species of non-fermentative and oxidase-positive gram-negative bacilli. (Rhoads *et al.*, 1995).⁽¹⁰⁾

Pictures from the (13-22) demonstrate micro organisms that have been selected from each sample to be definite, as well as the morphological characteristics of each micro organism in terms of color, the shape of a bacterial colony.



Picture (13) describes the shape of micro organism (6-1-A), (6-1-B)



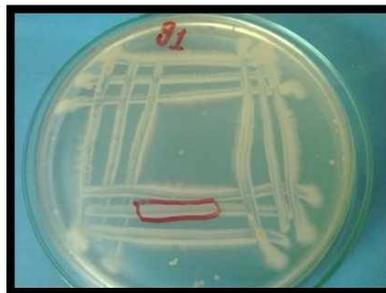
Picture (14) describes the shape of micro organism (1-2)



Picture (15) describes the shape of micro organism (1-1)



Picture (16) describes the shape of micro organism (10-1)



Picture (17) describes the shape of micro organism (9-1)



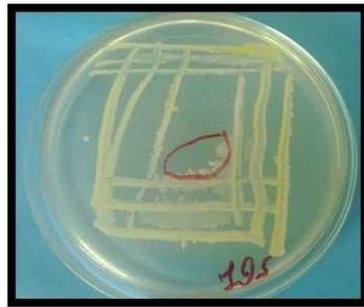
Picture (18) describes the shape of micro organism (6-1)



Picture (19) describes the shape of micro organism (19-2)



Picture (20) describes the shape of micro organism (18-1)



Picture (21) describes the shape of micro organism (19-5)



Picture (22) describes the shape of micro organism (19-4-A), (19-4-B)

II- Molecular Identification

a- Analysis of total cellular proteins

The total cellular proteins were separated and differentiated by sodium dodecylsulfate polyacrylamide gel electrophoresis (SDS-PAGE) as described by Laemmli (1970).⁽⁶⁾ In this technique proteins are separated according to their molecular masses and hence considered a fingerprint for each bacterial strain. The use of SDS (an anionic detergent) is a must because it denatures proteins and confers a negative charge to polypeptide in proportion to its length, then the polypeptide becomes negatively charged rods and are strongly attracted toward an anode (positively-charged electrode) in an electric field. The different steps of the procedure are detailed below.

1) Preparation of the bacterial proteins

Bacteria was grown in LB-broth (15 ml) at three different temperatures such as (37°C, 40°C and 42°C) with continuous shaking at 250 rpm. Cells were collected by centrifugation at 13,000 rpm for 5 minutes; the pellet was washed once with 1 M NaCl and twice with distilled water. The pellet was dissolved in an appropriate volume of distilled water according to its size. A 50µl of cell suspension was mixed with an equal volume of 2X treatment buffer, boiled for 90 sec in a water bath and immediately transferred to an ice-water bath. Just before loading onto the SDS-PAGE, a 2-3µl of tracking dye was added to each sample.

2) Preparation of SDS-polyacrylamide gel

The denatured gel consists of a stacking gel (4%) layer on top of a separating gel (12%) layer. All components of both layers of the SDS-PAGE, except APS, and TEMED, were mixed together in clean beaker and aerated under vacuum, then the polymerization initiator and cross linking solutions (APS, and TEMED) were added and swirled gently to avoid air bubbles. Then the separating gel solution was pipetted into vertical glass slab to about 2 cm of the top, a leveling thin layer of n-butanol was added on the top of the separating gel and then left to polymerize at room

temperature, for at least three hours. After polymerization, the n-butanol was poured off from the top of the separating gel and washed once with distilled water. The stacking gel solution was then pipetted on top of separating gel, the well-former comb was inserted and the gel was allowed to polymerize for another 2 hours at room temperature before use.

3) Separating cellular proteins

After complete polymerization, the gel was placed into the electrophoresis chamber filled with the tank buffer and the comb was slowly removed leaving the formed wells. The gel was run for 30 minutes at 100 volts before loading the protein samples to clean the wells from any residues. Using Hamilton syringe, an equal weight of the cellular proteins were loaded to each well. Protein marker was loaded into a separate well for calculating the molecular masses of proteins. The electrophoresis was run at 50 volts until protein samples emerged from stacking gel, then the power was raised to 100 volts until the tracking dye was close to run off the bottom of the gel. Then the power supply was turned off and the glass plate sandwich containing the gel was removed from apparatus and dismantled to stain the gel.

4) Staining of cellular proteins and visualization

The gel was stained by soaking in Coomassie Brilliant Blue R-250 stain solution and kept shaken for at least 6 hours at 37°C, 40 rpm. The gel was then removed from stain solution, washed with water and destained by solution (I) for at least two hours and solution (II) for one hour respectively. Protein bands can be seen by naked eyes, and the gel was photographed for analysis.

RESULTS:

Table (1) illustrates the positive samples for the initial tests of bacteria ratios, in accordance with the different stages of gown's use, we find (6) samples from total positive samples, representing 37.5 % of the positive samples in the pre-use, and other (6) samples representing (37.5%) of the

positive samples after surgery, and (4) samples | representing (25%) in the phase after sterilization.

Table (1) shows the positive samples for initial tests in accordance with the stages of gown's use

Samples type in accordance with the stages of use	positive samples	number of samples	Percentage
Positive samples at the stage (before use)	(1) - (7) - (10) - (13) - (14) - (17)	6	37.5%
Positive samples at the stage (after use)	(5) - (8) - (11) - (15) - (18) - (20)	6	37.5%
Positive samples at the stage (after sterilization)	(6) - (9) - (16) - (19)	4	25%

Classical and Biochemical identification of the bacterium

The isolated bacterium was found to be a rod-shaped, Gram negative organism. Biochemically, the bacterium isolated characterized by being negative for: *arabinose*, *maltose*, *adonitol*, *urea*, *oxidase* test, *tartrate* test, *lysine*, *acetamide*, *cetrimide*, *nitrate reductase*, *inositol*, production of *indole* and *hydrogen sulfide*, cannot hydrolyze *esculine*, reduces *nitrate*, or deaminate *tyrptophane* Table (1), (2), (3) as compared to the series of biochemical reactions and a large data base for

Gram negative of the MicroScanWalkAway -90 System.

Accordingly, the bacterium was identified as *Enterobacter cloacae*, a member of the *Enterobacteriaceae* family in accordance with their description in Bergey's Manual of Systematic Bacteriology (Williams, 1994). This test is the simplest to monitor the emergence of drug-resistant among organism not previously detected either within an individual health care facility or community.

Table (2): Biochemical test API20 (Identification of isolated Gram negative organism by automated MicroScanWalkAway-90 Microbiology System for samples {(10-1), (18-1), (19-4-A)})

Sample No.	10	18	19
Isolated organism	(1)	(1)	(4-A)
Bacteria name	<i>Y.pseudotp</i>	<i>P.luteola</i>	<i>P.vulgaris</i>
Biochemical Results	GLU	+	-
	INO	-	-
	URE	-	-
	LYS	-	-
	SCU	+	-
	MAL	-	-
	TAR	-	-
	ARA	-	-

Table (2) show that the bacteria which appeared was three types, the first is *Pseudomonas luteola* which is an opportunistic pathogen, found ubiquitously in damp environments, the second type was *Yersinia pseudotuberculosis* and it is a Gram-negative bacterium that causes Far East scarlet-like fever in humans, who occasionally get infected zoonotically, most often through the food-borne route, the third type was the bacteria *P. vulgaris*.

All three these bacteria types also can be resistant to some drugs and antibiotics such as *Cefuroxime*, and *Aztreonam*, and some of them can't resist some others such as *Ertapenem*, *Pip/Tazo*, and *Piperacillin*.

Also results showed that isolated bacteria does not has the potential to interact with a lot of sugars and organic compounds such as *Arabinose*, *Maltose*, *Urea*, *Uxidase*, *Lysine*, *Cetrimide*, *Acetamide* and *Tartrate test*.

Table (3): Biochemical test API20 (Identification of isolated Gram negative organism by automated MicroScanWalkAway-90 Microbiology System for samples {(1-2), (19-2), (19-5)})

Sample No.	1	19	19
Isolated organism	(2)	(2)	(5)
Bacteria name	<i>S.hominis-novo</i>	<i>S.cohnii- cohnii</i>	<i>S.capitis-ureo</i>
Biochemical Results	CV	+	+
	NOV	+	+
	URE	-	-
	LAC	+	-

	PYR	-	-	-
	TRE	-	-	-
	NACL	+	+	+
	ARA	-	-	-

Table (3) show that micro-organisms which appeared were a Gram positive, coagulase-negative member of the bacterialgenus Staphylococcus consisting of clustered cocci. the species commonly lives on human skin.

This Gram positive coccus is found as singles, pairs, and less frequently as irregular, grape-like clusters. Cells are catalase positive and exhibit facultatively anaerobic metabolism. Able to use glucose oxidatively and fermentatively. Salt tolerant in media with NaCl concentrations up to

10%. Growth occurs at temperatures between 15-40 C, with optimum growth occurring between 30-35 C. Isolated most frequently from human skin and infrequently from primates. No known involvement in human disease has been reported. Also results showed that isolated bacteria has the potential to interact with some sugars and organic compounds, but does not has the potential to interact with the few of these compounds such as *Arabinose* and *Urea*.

Table (4): Biochemical test API20 (Identification of isolated Gram negative organism by automated MicroScanWalkAway-90 Microbiology System for samples {(1-1), (6-1-A), (6-1-B)}, (6-2), (9-1), (19-4-B)}

Sample No.	1	6	6	6	9	19
Isolated organism	(1)	(1-A)	(1-B)	(2)	(1)	(4-B)
Bacteria name	<i>P.vulgaris</i>					
Biochemical Results	GLU	+	+	+	+	+
	INO	-	-	-	-	-
	URE	-	-	-	-	-
	LYS	-	-	-	-	-
	SCU	+	+	+	+	+
	MAL	-	-	-	-	-
	TAR	-	-	-	-	-
	ARA	-	-	-	-	-

Table (4) show that *Proteus vulgaris* is a rod-shaped, nitrate-reducing, indole+ and catalase-positive, hydrogen sulfide-producing, Gram-negative bacterium that inhabits the intestinal tracts of humans and animals. It can be found in soil, water, and fecal matter. It is grouped with the Enterobacteriaceae and is an opportunistic pathogen of humans. It is known to cause urinary tract infections and wound infections.

P. vulgaris can be resistant to some drugs and antibiotics such as *Cefuroxime*, *Aztreonam*, and *Amp/Sulbactam*, but it can't resist some others such as *Piperacillin*, *Pip/Tazo*, and *Ertapenem*, also results showed that *P. vulgaris* does not has

the potential to interact with some sugars and organic compounds such as *Arabinose*, *Maltose*, *Urea*, *Lysine*, and *Tartrate*.

Antibiogram (antibiotic sensitivity) of the bacterium:

The antibiotic sensitivity results suggested that only the genes encode for the antibiotics: piperacillin, Pip/Tazo, Imipenem, Ertapenem, Cefuroxime, Ceftazidime/K Clavulanate, Aztreonam and Amp/Sulbactam are carried on plasmids while the rest of the tested antibiotics are chromosomal encoded Table (4), (5), (6). This consistent with the disappearance of the two plasmids at 40 and 42°C.

Table (5):The antibiotic sensitivity of the bacterial isolates as done automatically by MicroScanWalkAway -90 Microbiology System for samples {(10-1), (18-1), (19-4-A)}

Sample No.	10	18	19
Isolated organism	(1)	(1)	(4-A)
Bacteria name	<i>Y.pseudotp</i>	<i>P.luteola</i>	<i>P.vulgaris</i>
Antibiotic sensitivity	<i>Amp/Sulbactam</i>	S	----
	<i>Aztreonam</i>	R	R
	<i>Ceftazidime/K Clavulanate</i>	----	----
	<i>Cefuroxime</i>	R	----
	<i>Ertapenem</i>	S	----
	<i>Pip/Tazo</i>	S	S
	<i>Piperacillin</i>	S	S

S= Susceptible I= Intermediate
R=Resistant --- = Not tested

Table (5) show that the bacteria which appeared were three types, the first is *Pseudomonas luteola* which is an opportunistic pathogen, found ubiquitously in damp environments. Originally designated in the *Chryseomonas* genus, the species has since been reassigned to the *Pseudomonas* genus.

The second type was *Yersinia pseudotuberculosis* and it is a Gram-negative bacterium that causes Far East scarlet-like fever in humans, who

occasionally get infected zoonotically, most often through the food-borne route.^[1] Animals are also infected by *Y. pseudotuberculosis*. The bacterium is urease positive, and the third type was the bacteria *P. vulgaris* which has been talked about in advance as shown in table (3).

All three these bacteria types also can be resistant to some drugs and antibiotics such as *Cefuroxime*, and *Aztreonam*, and some of them can't resist some others such as *Ertapenem*, *Pip/Tazo*, and *Piperacillin*.

Table (6): The antibiotic sensitivity of the bacterial isolates as done automatically by MicroScanWalkAway -90 Microbiology System for samples {(1-2), (19-2), (19-5)}

Sample No.	1	19	19
Isolated organism	(2)	(2)	(5)
Bacteria name	<i>S.hominis-novo</i>	<i>S.cohnii- cohnii</i>	<i>S.capitis-ureo</i>
Antibiotic sensitivity	<i>Amp/Sulbactam</i>	R	R
	<i>Ampicillin</i>	BLAC	BLAC
	<i>Cefuroxime</i>	R	R
	<i>Erythromycin</i>	R	R
	<i>Penicillin</i>	BLAC	BLAC
	<i>Tetracycline</i>	S	S
	<i>Vancomycin</i>	R	R

S= Susceptible I= Intermediate R=Resistant
BLAC= Beta-Lactamase Positive --- = Not tested

Table (6) show that micro-organisms which appeared were a Gram positive, coagulase-negative member of the bacterialgenus *Staphylococcus* consisting of clustered cocci. the species commonly lives on human skin; clinical isolates have shown high levels of antibiotic resistance.

A strain of *S. cohnii* and *S.hominis-novo* was found to contain a mobile genetic element very similar to the SCCmec (staphylococcal cassette chromosome encoding methicillin resistance) element seen in *Staphylococcus aureus*.

This Gram positive coccus is found as singles,

pairs, and less frequently as irregular, grape-like clusters. Cells are catalase positive and exhibit facultatively anaerobic metabolism. Able to use glucose oxidatively and fermentatively. Salt tolerant in media with NaCl concentrations up to 10%. Growth occurs at temperatures between 15-40 C, with optimum growth occurring between 30-35 C. Isolated most frequently from human skin and infrequently from primates. No known involvement in human disease has been reported, they also can be resistant to some drugs and antibiotics such as *Ampicillin*, *Cefuroxime*, *Erythromycin*, and *Vancomycin*.

Table (7): The antibiotic sensitivity of the bacterial isolates as done automatically by MicroScanWalkAway -90 Microbiology System for samples {(1-1), (6-1-A), (6-1-B)}, (6-2), (9-1), (19-4-B)}

Sample No.	1	6	6	6	9	19
Isolated organism	(1)	(1-A)	(1-B)	(2)	(1)	(4-B)
Bacteria name	<i>P.vulgaris</i>					
Antibiotic sensitivity	<i>Amp/Sulbactam</i>	R	R	R	R	R
	<i>Aztreonam</i>	R	R	R	R	R
	<i>Ceftazidime/K Clavulanate</i>	----	----	----	----	----
	<i>Cefuroxime</i>	R	R	R	R	R
	<i>Ertapenem</i>	S	S	S	S	S
	<i>Pip/Tazo</i>	S	S	S	S	S
	<i>piperacillin</i>	S	S	S	S	S

S= Susceptible I= Intermediate R=Resistant --- = Not tested

Table (7) show that *Proteus vulgaris* is a rod-shaped, nitrate-reducing, indole+ and catalase-



positive, hydrogen sulfide-producing, Gram-negative bacterium that inhabits the intestinal tracts of humans and animals. It can be found in soil, water, and fecal matter. It is grouped with the Enterobacteriaceae and is an opportunistic pathogen of humans. It is known to cause urinary tract infections and wound infections.

According to laboratory fermentation tests, *P. vulgaris* ferments glucose and amygdalin, but does not ferment mannitol or lactose. *P. vulgaris* also tests positive for the methyl red (mixed acid fermentation) test and is also an extremely motile organism.

When *P. vulgaris* is tested using the API 20E identification system it produces positive results for sulfur reduction, urease production, tryptophan deaminase production, indole production, sometimes positive gelatinase activity, and saccharose fermentation, and negative results for the remainder of the tests on the testing strip.

P. vulgaris can be resistant to some drugs and

antibiotics such as Cefuroxime, Aztreonam, and Amp/Sulbactam, but it can't resist some others such as Piperacillin, Pip/Tazo, and Ertapenem.

Molecular characterization of bacterial isolates:

Molecular characterization of any living organisms, including bacteria, depends on its own genome (chromosome and plasmid) and its expression products (proteins). Fractionation of the bacterial proteins shows the differences among these organisms.

1-Protein banding pattern of bacterial isolates:

The protein banding pattern of the six bacterial isolates are shown in Fig (2). The figure also includes the protein banding patterns of the Prestained protein ladder, Broad Range (10-230 kDa, BioLabs). Careful examination of these patterns revealed two distinctive protein banding patterns. Patterns one for isolates 1, 2 and 3 and pattern two for isolates 4, 5 and 6. A single band (as indicated by the arrow) distinguishes the two patterns.

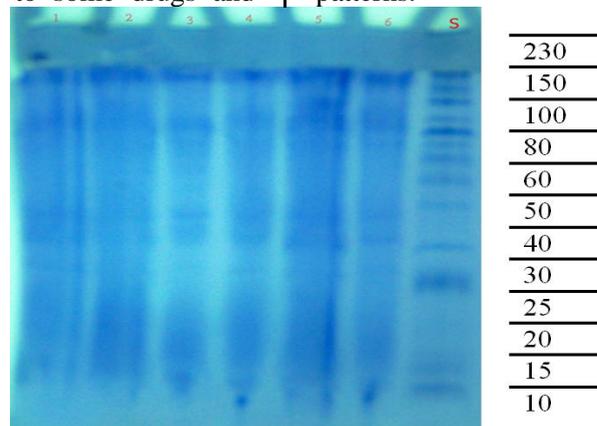


Figure (2) shows SDS-PAGE analysis of total cellular proteins of bacterial isolated (lanes from 1-6) from physician's coats, Mansoura Hospitals. S: Prestained protein ladder, Broad Range (10-230 kDa, BioLabs). The molecular weights of the ladder arranged from bottom up (as indicated by the arrows) are: 10, 15, 20, 25, 30, 40, 50, 60, 80, 100, 150, and 230 kDa bands.

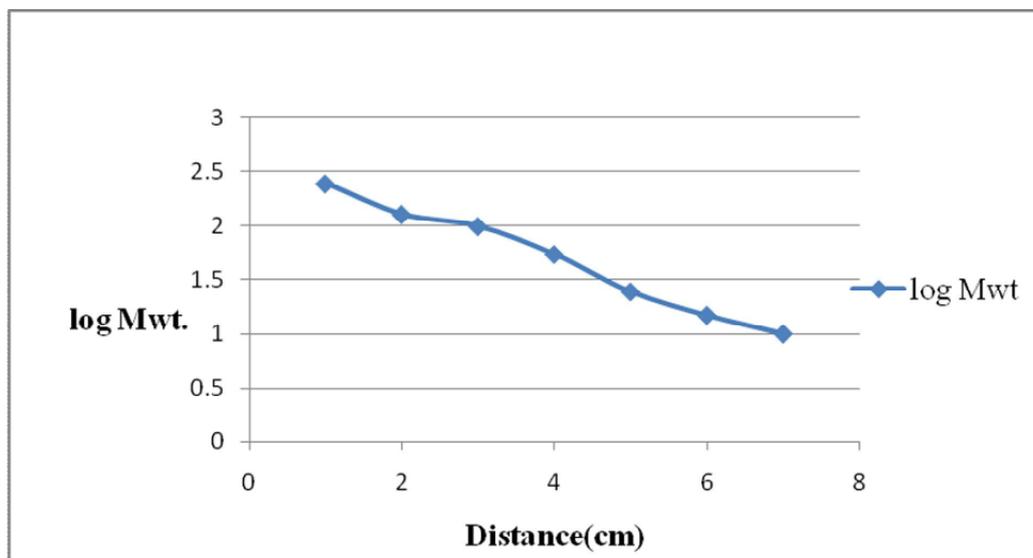


Figure (3): Standard curve of protein gel. Relation between migration distance of the standard protein bands and its log molecular weight.

A correlation between the migration distance of each protein band in the SDS_PAGE gel is calculated at a logarithmic scale of its molecular weight. The actual molecular weight is then calculated by taking the antilog for each band. The molecular weight is the scale to get the weight of each protein in the bacterial cell.

Discussion:

Firstly there is statistically significant relationship between the medical dressing used for operating rooms and injuring their wearers with some damages, and this is verified by conducting detect the presence of certain bacteria types that may be present dressed in operating rooms, tests, where the number of positive samples for initial tests (16 samples), including it represents the percentage (80%) of the total study sample of, as shown in figure (1).

This result also confirmed through what was confirmed by the results of the definition of bacteria which were appeared in the samples using a device (Micro Scan WalkAway - 90 Systems), which showed the presence of different strains of the two types of Gram Negative Bacteria, and Gram Positive Bacteria and these strains of which has the property of drug resistance, and that as indicated in tables (2), (3), (4), and these bacteria are often a clear impact on the health its wearer, it can be a reason for the transfer of many diseases such as skin infections, colorful tinea, and rashes, ringworm of the foot, and bran venous, breast inflammation, fever bedding, pneumonia, and this is consistent with what came from the results of the study (Nagda Mady: 1999)⁽⁹⁾, which confirmed that the clothes could represent a distinctive environment for the growth of organisms minute and special types of bacteria from Gram Negative, Gram Positive, Staphylococcus Aureus, as well as some types of fungi, especially given the lack of awareness by consumers of these clothes the best methods to deal with it and take care of her.

This is also consistent with the results of a study (Avi A. Weinbroum, et al.,) (2007)⁽¹⁾, which showed that more than 53% of anesthesiologists and surgeons do not respect any rules to wear surgical clothes outside the operating room, and (80%) of the surgeons who leaving the operating room remain to wear surgery for a long time after the end of the operations, thus providing opportunities for the transmission of these to others, and this result is also consistent with what came from the results of a study (J.A Wilson et al., 2007), which showed that 68% of nurses operating rooms' clothes loaded with bacteria types *Staphylococcus aureus*, and bacteria

Clostridium difficile, and that more than 50% of the nursing staff resort to disinfect their operation' clothing in foreign laundries, which cause an increase in its carry microbial, and which confirms the lack of awareness of these best methods to deal with these kinds of clothes.

Secondly: There is statistically significant relationship between the level of transactions conducted on the medical dressing used for operating rooms and maintain the health of their wearers, and this is what has been verified through as confirmed by the results of Table (1), which confirms the presence of bacteria in study samples in various stages (gowns before using for the first time - gowns after surgery - gowns after sterilization), we find the percentage (37.5%) of the study sample carrying the bacteria, before use for the first time, this gives an important indication of the importance of dealing with operating rooms' clothes and regarded as one of infection factors even before using, and these results confirmed by field visits to hospitals (the study sample), which made it clear that there is only one hospital in these hospitals is disinfected surgical operating rooms' gowns before being handed over to the surgeon for the first time, the (Internal Medicine Specialist Hospital), this represents a only 10% of hospitals (the study sample), and this percentage can be described very low, as evidenced by the tables numbers (5), (6), (7) that the bacteria isolated from the samples (1), (10) (samples at the stage of before using for the first time) can resistance drugs and antibiotics, which shows the seriousness of these bacteria, and this is consistent with the result of the study (Yonit Wiener et al., 2011)⁽¹¹⁾, which found that 60% of the hospital staff's clothes loaded with bacteria pathogens including drug-resistant bacteria, and these clothes play an important role in the transmission of infection within these hospitals.

While the results are also shown in Table (1) that there is a (25%) of the positive study samples loaded with bacteria at some point (after sterilization), and these bacteria, for example, found samples numbers (6), (9), (16), (19), as shown in the tables numbers (5), (6), (7), and these bacteria also have the ability to resist antibiotics and other pharmaceuticals, which indicates the inefficiency of sterilization processes that occur in hospitals (the study sample), thereby increasing the microbial load wear operating rooms before re-use in other operations, so we can say that the level of transactions conducted on the surgical dressing rooms is not the optimum level, which increases the chances of the spread of infection among workers in the medical sector as well as

undecided for patients.

Conclusion:

1. Medical clothes represent a distinctive environment for the growth of micro-organisms, particularly bacteria species of Gram Negative, Gram Positive, in light the lack of awareness of doctors and nursing staff with the best methods to deal with, as well as lack of awareness of those who care for it.
2. The level of transactions conducted on the surgical dressing rooms is not the optimum level, which increases the chances of the infection spread among workers in the medical sector as well as for patients.
3. Washing and sterilization operations are consider the most prevalent ways in the hospitals to take care of operating rooms' clothes, and these ways affect largely on their appearance and their consumption age, and that this method represents a burden on the budget of these hospitals, and these way from the viewpoint of its users are not enough to eliminate what may afford these clothes from microbes.

Recommendations:

1. Trend towards the use of disposable surgical operating rooms' clothes in hospitals due to their high level of health safety and their low cost compared to durable surgical operating rooms' clothes.
2. Prohibit the use of operating rooms clothes for the first time before adequately sterilized.
3. Develop standardized and safe controls to get rid of operating rooms clothes in order to achieve environmental safety.
4. A lot of comparative studies should be applied on disposable and durable surgical operating rooms' clothes.

References:

1. Avi A. Weinbroum, et al., (2007): Wearing surgical attire outside the operating room (A survey of habits of anesthesiologists and surgeons in Israel, *Journal of the American College of Surgeons*, 205 (2): 314- 318.
2. C.L. Shelton, et al., (2010): Can changes in clinical attire reduce likelihood of cross-infection without jeopardizing the doctor-

patient relationship?, *Journal of Hospital Infection*, 74 (1): 22-29.

3. Collee, J. G., et al., (1996): *Practical Medical Microbiology*, 14th ed, Produced by Longman Singapore Publishers (pte) Ltd:385.
4. Fatma Ali Metwaly (2003): *Quality system assessment for the operations of clothing design*, Ph.D. Thesis, College of Applied Arts, Helwan University.
5. Fawzia Abd El salaam (2012): *The possibility of reaching some functional standards for woven and non-woven products to develop doctor's uniform in the operating room*, Master Thesis, College of Home Economics, Menoufiya University.
6. Laemmli UK (1970): Cleavage of structural proteins during the assembly of the head of bacteriophage T4, *Nature* 227, 680-685.
7. Marisha Burden, et al., (2011): *Newly Cleaned Physician Uniforms and Infrequently Washed White Coats Have Similar Rates of Bacterial Contamination After an 8-Hour*, *Journal of Hospital Medicine*.
8. Mona Samir Fattouh (2010): *The possibility of reaching appropriate clothing for patients with eczema structural as one of the affected skin diseases for patients of costumed children*, Master Thesis, College of Home Economics, University Menoufia.
9. Nagda Mady (1999): *Adverse effects on human health resulting from the use of certain textile products and Ghazlah and the role of home economics curriculum in raising health awareness*, Master Thesis, Faculty of Home Economics, Menoufiya University.
10. Rhoads S, et al., (1995): *Comparison of MicroScan WalkAway System and Vitek System for identification of gram-negative bacteria*, *Journal Clin Microbiol*, 33: 3044-3046.
11. Yonit Wiener-Well, et al., (2011): *Nursing and physician attire as possible of nosocomial infections*, *American Journal of Infection Control*, 39 (7): 555-559.

* References (4),(5),(8),(9) in Arabic