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## **Antibacterial Synergistic Activity of Ofloxacin and Ornidazole Treated Biomedical Fabrics against Nosocomial Pathogens**

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### **ABSTRACT**

Microbial damage of fabrics and microbial-contaminated fabrics are known to be the major source of nosocomial cross-infections in hospitals. The hospital fabrics possess susceptible surface properties to harbour diverse group of bacteria. The main aim of this study is to treat the fabric materials using two groups of antibacterial drug combinations based on their synergistic behaviour. Hence for the first time synergistic ofloxacin and ornidazole drugs were covalently bound to the textile materials using reactive dye method. Using the standard AATCC Test Method-100 the antibacterial activity and durability of drug treated textile materials was evaluated before and after wash. Drug treated textile materials (before wash) showed maximum percentage of reduction with a reduction percentage of 90% for *E. coli* (nylon) and 92.85% for *S. aureus* (polyester). After 5th wash, the treated textile materials showed maximum reduction percentage of 63.4% for *E. coli* (nylon) and 64.1% for *S. aureus* (nylon). The development of antibacterial textile finish in the study could have the possibility to eliminate the drug resistance properties of hospital based nosocomial pathogens.

**Key words:** Reactive dye method, AATCC-100, AATCC-124, beta-cyclodextrin, agarose gel electrophoresis

### **INTRODUCTION**

Health and hygiene are the primary requirements for human beings to live comfortably and work with maximum efficiency. Despite progress in public health and hospital care, infections continue to develop in hospitalized patients and healthcare workers. Nosocomial infections may be transmitted from various sources in the hospitals. One such source is dissemination of pathogens from biomedical products including textiles. Hospital textile materials such as theatre drapes, gowns, masks, sheets and pillowcases are known to be major sources of cross-infection. A major concern for healthcare workers is the problem of transmission of pathogens from these biomedical textile materials. This includes penetration of biological liquids (blood, body fluids) and associated bacteria into the hospital fabric materials used by healthcare workers and patients. Harmful pathogens in these fluids can reach and penetrate the skin of surgeons and/or patients, with an associated potential for infection. Secondly, bandage and wound dressing materials carrying both

aerobic and facultative anaerobic bacteria may also be disseminated among the patients and workers. The penetrating and transmitting organisms are *Escherichia coli*, methicillin resistant *Staphylococcus aureus*, *Klebsiella pneumoniae*, *S.epidermis* and anaerobic bacteria like *Bacteriodes fragilis*, *Peptostreptococcus* sp. (Elshafei and El-Zanfaly, 2011).

The effective strategy for reducing such nosocomial infections is to reduce the dose of microorganisms throughout the healthcare complex using antimicrobial technologies to treat the textile surfaces and to maintain the standard of hygiene. More recently, an awareness of general sanitation, contact disease transmission and personal protection have led to the development of antibacterial fibres (Curtis and Monticello, 2002). Most of these approaches entail the attachment of a biocidal or bacteriostatic agent to the fabric surface. The mechanisms used to attach these agents to the fabric include the layer deposition of silver nanoparticles onto fabric structures (Dubas *et al.*, 2006). Graft polymerization of N-halamide monomers onto cellulosic substrates (Liu and Sun, 2006). Addition of quaternary ammonium salts onto cotton fabrics using a covalently bound adducts (Son *et al.*, 2006). Co-valent attachment of a chloromelamine derivative (Sun *et al.*, 2005) and the attachment of chitosan to cotton fabric via cross-linking agents (El-tahlawy *et al.*, 2005; Ye *et al.*, 2006). The problem with straight forward antimicrobial loading into the fabric is the generation of resistance. Antimicrobial resistance has a significant impact on patient outcome by enhancing virulence, delaying the administration of appropriate therapy and subsequent recovery (Cosgrove and Carmeli, 2003).

Even though different type of antimicrobial agents were used to be more effective in antibacterial activity, still due to several above mentioned factors they are not considered as biocompatible. Hence, these problems led us to study the effects of introducing two different groups of synergistic antimicrobial drugs (a fluoroquinolone drug and a nitroimidazole drug) into the textile materials. The drugs were chemically converted in order to obtain a reactive dye type molecule based on the factor described by Saginur *et al.* (2006). They reported that the accepted clinical practice to treat biomedical-associated infections was the use of combination therapy in which two or more antimicrobials are blended at different combinations. So that broader spectrum of activity is achieved at a lower concentration resulting in more effective therapy and decreased resistance. The character of synergism mainly depends on the mode of action of a drug. Both fluoroquinolone and nitroimidazole drugs acts on the DNA of bacteria thus targeting the inhibition of DNA synthesis and replication.

Taking into consideration of the above facts, the present research work was designed with the objective of developing a process for rendering antimicrobial coating to the biomedical products. Also, the study aims to determine the effect of the cross-linking of antimicrobial agent to the product substrate on the antimicrobial efficacy, durability, persistence and comfort properties.

The objectives are as follows:

- To produce antimicrobial finished textile materials by reactive dye method
- To evaluate the antibacterial activity of finished textile materials by AATCC-100 method
- To investigate the durability of finished textile materials using AATCC-124 method

## MATERIALS AND METHODS

In the present research, testing the synergistic activity of antimicrobial drugs and effect of drugs on bacterial DNA tests were carried out in Microbiology laboratory, CMS College of Science and Commerce, Coimbatore, India, from January 2011 to February 2011. Reactive dye method and

Antibacterial activity of treated fabric materials (AATCC 100 method) were carried out in Microbiology laboratory, PSG College of Arts and Science, Coimbatore, India, in March 2011.

**Textile materials:** The fabric from a commercial producer used for various purposes in the healthcare centre was used as the test fabric. 100% polyester and 100% nylon was selected and sterilized prior experimentation. The fabric was cut into squares (swatches), approximately 5 cm×5 cm, before being treated. After treatment, the swatches were wrinkled removed and sterilized in prior to the antibacterial assay.

**Clinical isolates (90 S agitation method):** Clinical pathogens were isolated from the contaminated hospital used fabrics by the method described by Cody *et al.* (1984).

**Antibacterial drugs:** Medical grade fluoroquinolone compound (ofloxacin) and nitroimidazole compound (ornidazole) were purchased from Sigma chemical Co. The drugs were checked for their purity based on their specific wavelength using UV-VIS spectrophotometer.

**Cross-linker:** Food and medical grade  $\beta$ -cyclodextrin (Hi media) was used as the cross-linker between the textile materials and antibacterial drugs.

**Minimal Inhibitory Concentration (MIC):** The Minimal Inhibitory Concentration (MIC) of each drugs were determined by a standard agar dilution method as described by Qaziasgar and Kermanshahi (2008).

**Checker board titration method:** After determining the MIC of each drug, a checkerboard titration method as described by Qaziasgar and Kermanshahi (2008) was performed to investigate the synergistic activity of ofloxacin and ornidazole on each challenge bacteria using Fractional Inhibitory Concentration Index (FICI).

**Fractional inhibitory concentration index:** To determine the synergistic activity of the selected drugs on each challenge bacteria the Fractional Inhibitory Concentration Index (FICI) was calculated using the equation and interpretation described by Bharadwaj *et al.* (2003).

**Method of imparting antibacterial drugs on textile materials:** The selected textile materials were treated with synergistic antibacterial drugs using reactive dye method as described by Chun and Gamble (2007). Briefly, the reactive drugs were synthesized using the selected cross-linker and sodium hydroxide solution. The synthesized reactive drugs were then made imparted to each of the textile materials.

**Assay for antibacterial properties:** The drug treated textile materials (nylon and polyester) were subjected to evaluate the antibacterial activity on each challenge bacteria. The assay used for evaluating the antibacterial properties was based on the standard AATCC Test Method 100-1999 as described by Rajendran *et al.* (2010). The difference in number of viable bacteria was evaluated on the basis of the percentage reduction. Percentage reduction was calculated using the following formula.

$$R = (A-B) / A \times 100$$

where, R is percentage reduction, A is the number of bacteria in the broth inoculated with treated test fabric sample immediately after inoculation i.e., at zero contact time and B is the number of bacteria recovered from the broth inoculated with treated test fabric sample after the desired contact period-18 h.

**Wash fastness test:** The treated textile materials were analysed to investigate the durability of the drugs after undergoing periodical and consecutive washings. The treated textile materials were washed based on the standard AATCC Test Method-124 as described by Rajendran *et al.* (2010). The treated and control samples were washed 2 and 5 times, respectively.

**Effect of synergistic drugs on DNA:** The purified test cultures of *E. coli* and *S. aureus* were treated to each 200 µg of two antimicrobial compounds. Both treated and untreated cultures were incubated overnight at 37°C. Using a standard method of Sambrook and Russel (2001), DNA from both the treated and untreated cultures were extracted. Extracted DNA samples were analysed under UV light after running in 0.8% agarose gel electrophoresis column.

**Statistical analysis:** Using chi-square analysis, the hypothesis was selected as antimicrobial agent has good activity. The difference in the number of viable bacteria after '0' contact time and 18 h contact time were statistically calculated with p<0.05 considered significant.

## RESULTS

**Identification of clinical isolates:** The pathogens isolated and identified from the contaminated site of fabric by 90 s agitation method belong to different Gram-positive and Gram-negative bacteria. In Table 1, the cultural characteristics of one Gram-positive representative bacterium (*Staphylococcus aureus*) and one Gram-negative representative bacterium (*Escherichia coli*) were reported. *Staphylococcus aureus* and *Escherichia coli* were used throughout the study based on their significance of nosocomial characteristics.

**Synergistic activity of ofloxacin and ornidazole:** In Table 2 the strains of *Escherichia coli* and *Staphylococcus aureus* showed synergism (p<0.5) against ofloxacin and ornidazole when mean

Table 1: Identification of challenge bacteria

| Isolates                     | Cultural characteristics |           |     |
|------------------------------|--------------------------|-----------|-----|
|                              | TSB+5% blood             | MacConkey | EMB |
| <i>Staphylococcus aureus</i> | +                        | +         | –   |
| <i>Escherichia coli</i>      | –                        | +         | +   |

TSB+5% blood: Partial haemolytic with greenish zones for *S. aureus*. MacConkey: Lactose fermenting colonies for *S. aureus* and *E. coli*  
EMB: Green metallic sheen colonies

Table 2: Mean Fractional Inhibitory Concentration Index (FICI)

| Isolates                     | FICI | Interpretation |
|------------------------------|------|----------------|
| <i>Escherichia coli</i>      | 0.5  | Synergy        |
| <i>Staphylococcus aureus</i> | 0.5  | Synergy        |

Synergy: mean FICI<0.5

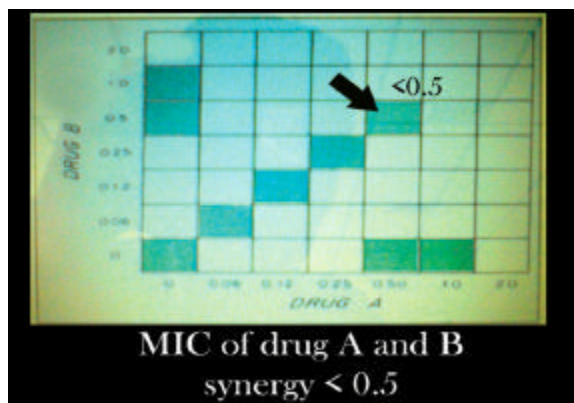


Fig. 1: Checker board titration method to evaluate the synergism between two drugs  
Diagrammatically illustrated the synergism between two drugs: ofloxacin-ornidazole  
(Figure extracted from Qaziasgar and Kermanshahi, 2008)

fractional inhibitory concentrations were calculated. A hand drawn model of checker board titration method was illustrated in Fig. 1 showing the inhibitory concentrations ( $p < 0.5$ ) of ofloxacin and ornidazole against the test organisms.

**Antibacterial activity of treated fabric material by reactive dye method:** The antibacterial activities of the fabric material (nylon, polyester) covalently bound with synthesized reactive synergistic drug combinations (ofloxacin-ornidazole) and cross-linker (beta-cyclodextrin) before and after wash (2, 5 times) were assayed by AATCC 100 method. In Table 3 the antibacterial activity of drug treated textile materials exposed for 18 hours (before wash) showed more antibacterial activity against *S. aureus*. When compared to the initial dose of organisms (0th time) only  $9 \text{ Cf} \times 10^3 \text{ mL}^{-1}$  for polyester materials were observed. After 5th wash, minimum of  $27 \text{ cfu} \times 10^3 \text{ mL}^{-1}$  were observed for nylon materials. Similarly in Table 4 the drug treated textile materials exposed for 18 h (before wash) showed more antibacterial activity against *E. coli*. When compared to the initial dose of organisms (0th time) only  $8 \text{ cfu} \times 10^3 \text{ mL}^{-1}$  for nylon materials were observed. After 5th wash only  $27 \text{ cfu} \times 10^3 \text{ mL}^{-1}$  and  $28 \text{ cfu} \times 10^3 \text{ mL}^{-1}$  were observed for nylon and polyester materials which indicate that the drugs were durable after consecutive washings.

**Percentage reduction of challenge bacteria:** Before wash and after wash textile materials (nylon, polyester) covalently bound with synthesized reactive drugs (ofloxacin-ornidazole) was assayed for antibacterial activity by AATCC 100 method. The difference in number of viable bacteria was evaluated on the basis of the percentage reduction. In Table 5 it was reported that the drug treated textile materials (before wash) showed maximum reduction percentage of 90 (%) for *E. coli* (nylon) and 93 (%) for *S. aureus* (polyester). After 5th wash, the treated textile materials showed maximum reduction percentage of 64 (%) for *S. aureus* (nylon) and 65 (%) for *E. coli* (polyester).

**Effect of synergistic drugs on bacterial DNA:** The exposure of bacterial pathogens, *S. aureus* and *E. coli* to ofloxacin-ornidazole was done to understand the interfering activity on the DNA

Table 3: Textiles treated with ofloxacin-ornidazole cross-linked to beta-cyclodextrin (*Staphylococcus aureus*)

| Fabric sample treated with ofloxacin-ornidazole                           | No. of colonies (cfu $\times 10^3$ mL $^{-1}$ ) |                                  |
|---|---|----------------------------------|
|   | Nylon+ $\beta$ -cyclodextrin                    | Polyester+ $\beta$ -cyclodextrin |
| Treated fabrics exposed with challenge bacteria at 0th time (before wash) | 91  | 96                               |
| Treated fabrics exposed with challenge bacteria after 18 h (before wash)  | 12  | 9                                |
| Treated fabrics exposed with challenge bacteria at 0th time (2nd wash)    | 82  | 80                               |
| Treated fabrics exposed with challenge bacteria after 18 h (2nd wash)     | 31  | 28                               |
| Treated fabrics exposed with challenge bacteria at 0th time (5th wash)    | 82  | 83                               |
| Treated fabrics exposed with challenge bacteria after 18 h (5th wash)     | 27  | 31                               |

Table 4: Textiles treated with ofloxacin-ornidazole cross-linked to beta-cyclodextrin (*Escherichia coli*)

| Fabric sample treated with ofloxacin-ornidazole                           | No. of colonies (Cfu $\times 10^3$ mL $^{-1}$ ) |                                  |
|---|---|----------------------------------|
|   | Nylon+ $\beta$ -cyclodextrin                    | Polyester+ $\beta$ -cyclodextrin |
| Treated fabrics exposed with challenge bacteria at 0th time (before wash) | 91  | 94                               |
| Treated fabrics exposed with challenge bacteria after 18 h (before wash)  | 8   | 10                               |
| Treated fabrics exposed with challenge bacteria at 0th time (2nd wash)    | 81  | 86                               |
| Treated fabrics exposed with challenge bacteria after 18 h (2nd wash)     | 32  | 35                               |
| Treated fabrics exposed with challenge bacteria at 0th time (5th wash)    | 84  | 82                               |
| Treated fabrics exposed with challenge bacteria after 18 h (5th wash)     | 27  | 28                               |

From the data (Table 3 and 4) the percentage reduction of challenge organisms against the treated fabrics were calculated and reported in Table 5

Table 5: Percentage reduction of challenge organisms against textile treated with synergistic reactive drugs (Before and after wash)

| Samples   | Reduction of bacteria (%) |          |          |                |          |          |
|---|---------------------------|----------|----------|----------------|----------|----------|
|   | <i>S. aureus</i>          |          |          | <i>E. coli</i> |          |          |
|   | Before wash               | 2nd wash | 5th wash | Before wash    | 2nd wash | 5th wash |
| Treated nylon exposed with challenge bacteria at 0th time     | 0                         | 0        | 0        | 0              | 0        | 0        |
| Treated nylon exposed with challenge bacteria after 18 h      | 88                        | 65       | 61       | 90             | 64       | 63       |
| Treated polyester exposed with challenge bacteria at 0th time | 0                         | 0        | 0        | 0              | 0        | 0        |
| Treated polyester exposed with challenge bacteria after 18 h  | 93                        | 67       | 64       | 88             | 68       | 65       |

Using chi-square parameter, the hypothesis was selected as antimicrobial agent has good activity against the challenge organisms. The degree of freedom obtained from the calculated value was  $V = 1$ . For all the data, the table value is greater than the calculated value. Hence, the difference in the number of viable bacteria after '0' contact time and 18 hours contact time were statistically calculated with  $p < 0.05$  considered significant. Effect of initial dose [0 contact time] of *Staphylococcus epidermidis* or *Escherichia coli* for reactive drugs (ofloxacin-ornidazole) on treated nylon fabrics when compared with final dose [18 hours contact time] for reactive drugs (ofloxacin-ornidazole) on treated nylon fabrics, showed that the numbers of viable bacteria in drug-treated fabric [18 hours contact time] ( $p < 0.05$ ) were less than the number of viable bacteria at 0 contact time of treated fabrics ( $p > 0.05$ ). Similar statistical result was obtained for the drug treated polyester fabric materials

synthesis of challenge organisms. The DNA extracted from the synergistic drug treated and untreated samples were subjected to agarose gel electrophoresis technique. In Fig. 2a and b the difference in the appearance of DNA bands under UV light indicates the difference of drug treated cells from drug untreated cells. The formed DNA band width, the intensity of illumination and UV absorption were more for the ethidium bromide stained DNA bands extracted from untreated than treated. Hence the result revealed the interference rendered by synergistic drugs on DNA synthesis mechanism of the pathogens at time of exposure.

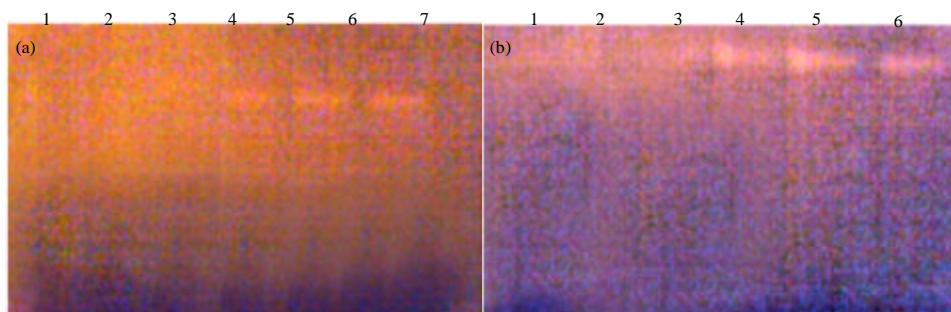


Fig. 2: Effect of ofloxacin and ornidazole on DNA of challenge bacteria *Escherichia coli* *Staphylococcus aureus*. (a) *Escherichia coli* and (b) *Staphylococcus aureus*. Lane 1, 2, 3- synergistic drug treated bacterial samples. Lane 4, 5, 6-untreated bacterial samples.

## DISCUSSION

Nosocomial infections or hospital acquired infections are diseases that develop during an admission to hospital and are a consequence of treatment, procedures of treatment or work of hospital staff. These infections are dangerous because they are caused by bacteria that are developed and transmitted within the hospital and hospital based products, where they may reach a high level of resistance to antibiotics (Maheswaran *et al.*, 2007). Mohammadi *et al.* (2010) evaluated urine cultures and the antimicrobial resistance patterns of *E. coli* detected from the hospitalized urine culture. The most resistance rates for *E. coli* detected from urine culture were to ampicillin with 98.4% and to amoxicillin with 83.7%, respectively. Their findings demonstrated the significance of resistance increase of *E. coli* detected from urine culture to various groups of antibiotic drugs, caused by the irregular use of antibiotics. Bratu and Quale (2006) reported that carbapenem antibiotics are typically reserved for serious nosocomial infections. Several classes of  $\beta$ -lactamases have emerged that possess carbapenem-hydrolyzing activity. Most nosocomial pathogens that possess a carbapenem-hydrolyzing  $\beta$ -lactamases are frequently resistant to other classes of antibiotics, including aminoglycosides and fluoroquinolones. Nawaz *et al.* (2001) reported, fluoroquinolones are broad spectrum synthetic antibiotics and are used for the treatment of many types of systematic infections. They analysed forty nine commercial pharmaceutical formulations including Ciprofloxacin, Norfloxacin Ofloxacin and Pefloxacin using UV and colorimetric methods for their active ingredient. The amount of fluoroquinolone differs in the range of +12 to -11% reporting that these amounts lie in the therapeutic window and without affecting their therapeutic action. Based on the mode of action of different groups of above mentioned drugs on bacterial components, different combinations of synergistic drugs were selected for the present study. The combinations of drugs were selected based on the following suggestions proposed by Bharadwaj *et al.* (2003), Shrivastava *et al.* (2009), Elayarajah *et al.* (2011) and Jasmine *et al.* (2007). According to Bharadwaj *et al.* (2003), combination of a quinolone drug with a nitroimidazole drug enhances the antimicrobial properties against both aerobic and anaerobic bacterial infections. Shrivastava *et al.* (2009) evaluated the antimicrobial properties of a new Fixed Dose Combination (FDC) of Ofloxacin-Ornidazole for infusion against some aerobic bacteria in comparison with Ofloxacin and Ornidazole individually. They reported that the MIC value of FDC of Ofloxacin-Ornidazole was higher than both Ofloxacin and Ornidazole. Their data suggests that



fixed dose combination of Ofloxacin and Ornidazole can be a good option for use in mixed microbial infection of aerobic bacteria, anaerobic bacteria and pathogenic protozoans. Elayarajah *et al.* (2011) indicated the synergistic effect of norfloxacin and metronidazole to prevent the ureteral stent associated infections. Jasmine *et al.* (2007) reported the minimal inhibitory concentration of  $\beta$ -lactam antibiotics to evaluate its synergistic interaction with medicinal plant extractions.

The synergistic ofloxacin and ornidazole were made reactive using the reactive dye method to bind covalently on the textile materials. Reactive dye method was used in the textile industries to impart dyes on cotton, polyester and other fabrics. Similar reactive dye method was used by Chun and Gamble (2007) to covalently bind the two synergistic drugs, trimethoprim and sulfamethoxazole on cotton fabrics. Several studies were made using reactive dye method for various applications in textile industries. Najafi *et al.* (2008) used reactive dyestuff to improve the adhesion of chitin to the surface of polyester/cotton fibres. The dyed samples showed good rubbing and washing colour fastness properties within the range of colour change. The colour strength of the dyed samples increased with the increased deposition of chitin on the fabric. Ahmad *et al.* (2001) compared the shade matching and wash fastness of vat dyed and reactive dyed cotton and polyester fabrics. Their study revealed that the reactive dyes might be suitable substitutes of vat dyes. Mostafa and Samarkandy (2005) used poly methacrylonitrile (MAN)-pregelled starch graft copolymers as a new thickener for printing cotton with reactive dyes. They also evidenced that the reactive dye added grafted polymer has the properties of providing excellent colour strength as well as overall fastness.

B-cyclodextrin was used as the cross-linker between the textile surface (nylon, polyester) and synergistic drugs (Ofloxacin-Ornidazole). Similar work was carried out by Chun and Gamble (2007) using cyanuric chloride as cross-linker between cotton and the antimicrobial - drugs (trimethoprim sulfamethoxazole). Szejtli (2003) reported that in the future, cyclodextrins might play a significant role in the textile industry to remove surfactants from washed textiles. Also he stated that when cyclodextrins bound chemically to fibers, it enhances hydrophilicity and inclusion complex forming ability to immobilize insect repellents and antimicrobial agents. Baboota *et al.* (2003) reported that the cyclodextrins (CDs) have been found as potential candidates because of their ability to alter physical, chemical and biological properties of guest molecules through the formation of inclusion complexes.

The antimicrobial activities for all the treated fabrics were determined based on AATCC-100 method as described by Rajendran *et al.* (2010) (before and after wash – AATCC-124 method). The reduction percentage of *S. aureus*, *E. coli* in the nylon fabric treated with reactive ofloxacin-ornidazole (before washing) was 88 (%) and 90 (%). The reduction percentage of *S. aureus*, *E. coli* in the polyester fabric treated with reactive ofloxacin-ornidazole (before washing) was 93 (%) and 88 (%). Similar result was observed in the study conducted by Chun and Gamble (2007) using trimethoprim and sulfamethoxazole treated cotton fabric samples cross-linked with cyanuric chloride. Their results indicate that both trimethoprim and sulfamethoxazole depressed the bacterial density of *K. pneumonia* and *S. aureus* significantly after 24 h incubation. The obtained values in our research are in support to the above mentioned reviews.

The swatches of treated and untreated nylon fabric were washed 2 and 5 times to determine if the antibiotic binding to the fabric would be durable through normal washing. The reduction percentage of *S. aureus*, *E. coli* in the nylon fabric treated with reactive ofloxacin-ornidazole (after 5th wash) was 61 and 63%. The reduction percentage of *S. aureus*, *E. coli* in the polyester

fabric treated with reactive ofloxacin-ornidazole (after 5th wash) was 64 and 65%. Similar wash-fastness method was described by Sathianarayanan *et al.* (2010). In their work an ecofriendly natural antibacterial finish has been prepared from the plant extracts for textile application. Herbal extracts from *Ocimum sanctum* (tulsi leaf) and rind of *Punica granatum* (pomegranate) have been applied to cotton fabric by the method of direct application, micro-encapsulation, resin cross-linking and their combinations. Except the method of direct application, all other treatments show good washing durability up to 15 washes. In another study conducted by Chun and Gamble (2007) using pure different synthetic reactive chemicals, trimethoprim and sulfamethoxazole, both unwashed and washed treated fabrics had significantly lower bacterial density than the untreated fabric and the averages were not significantly different among the three treated fabric. This result shows that the treated nylon fabric displayed antibacterial properties that persisted through 10 laundering. Rattanawaleedirojn *et al.* (2008) reported that antibacterial efficacy of nanosilver finished fabrics on *Staphylococcus aureus*. Bacteriological tests were performed against *S. aureus* as model for gram positive bacteria. The result indicates that the percentage reduction of bacteria in treated fabric was less than 99.9%. Also, the *S. aureus* was completely attracted on the silver finished textile even after being exposed to 20 consecutive hand laundering condition. The obtained values in our research are in support to the above mentioned reviews.

The findings in our study agree well with the experimental data reported by Chun and Gamble (2007), Sathianarayanan *et al.* (2010) and Rattanawaleedirojn *et al.* (2008) suggesting that the two synergistic drug combinations as reactive drugs can covalently bind to all fabric materials and inhibit the growth of both Gram-positive and Gram-negative bacterial organisms before wash and after wash. With controlled parameters the efficiency of the drugs in inhibiting the organisms can be increased even after 10 consecutive washings. The obtained values in our research are in support to the above mentioned reviews.

The formed DNA band width, the intensity of illumination and UV absorption were more for the ethidium bromide stained DNA of the untreated cultures than the antibiotic treated. The test result reveals the inhibition of DNA synthesis was mainly induced by the synergistic effect of both ofloxacin and ornidazole. The obtained results were compared with the report stated by Dollery (1999). In his study he stated that ofloxacin inhibits the activity of one of the A-subunits of the bacterial enzyme DNA gyrase which is responsible for the negative super coiling of DNA and an essential conformation for DNA replication. Similar mode of action of ornidazole was observed in the study conducted by Kucers *et al.* (1997). They analysed that 5-nitro group of ornidazole undergoes inhibition of DNA synthesis and the reduced metabolite causing a loss of the DNA helical structure with subsequent DNA strand breakage.

## CONCLUSION

The treated fabric displayed antibacterial properties that persisted through 5 consecutive laundings. Since ofloxacin, ornidazole were both easily prepared to act as reactive dyes, most of other commonly known antimicrobial compounds having the similar reactive sites may be used in a similar manner and future research should be expanded to include testing a wide spectrum of antimicrobial compounds. The ease of application may extend to the use of other antimicrobial drugs to provide value to textile fabrics where tailored antibacterial fabric is desired.

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