Invitro Antibacterial Activity Of Oxidized Regenerated Cellulose (Surgi ORC®) Against Antibiotic Resistance And Nosocomial Pathogens In Post Operative Surgical Site Infections (PSSI)

Vidya Sagar¹, Piyush Patel², Avini Rana³, Bhavin Trivedi⁴, Deepak Patel⁵

¹Head – R&D / QA& RA, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213
²Manager- QC, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213
³Assist Manager – RA, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213
⁴Assist Manager - QA & RA, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213
⁵Head – Production, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213

ARTICLE INFO

Abstract

The continued sustained progress of antibiotic-resistant microorganisms and nosocomial pathogens creates a critical problem for patients undergoing surgical procedures and specifically at post-operative surgical sites. In the event of post-operative infections caused by these microorganism, treatment and recovery for the patient becomes problematic. Hemostats are effective in control of bleeding with different mechanisms of action at various surgical sites. Oxidized Regenerated Cellulose (ORC) is an effective passive haemostat and has a proven antibacterial activity attributed to its low pH. The present study reports the invitro antibacterial activity of a commercial brand of ORC (Surgi ORC, Aegis Lifesciences, India) on antibiotic resistance and nosocomial pathogens. Thirty-one microorganisms of ATCC strains were studied by microbial challenge test on recovery plates on four variants of Surgi ORC. All the variants showed significant antibacterial activity on six of seven strains of antibiotic and Multi Drug Resistance (MDR) microbes. Surgi ORC presented mild activity only on MDR Clostridium difficile. Of the 24 non-antibiotic resistant and other pathogens tested, Surgi ORC showed extremely significant microbial activity in 13 strains (P<0.001), very significant and moderate activity in 7 strains (P<0.01) and mild activity in only 4 strains of microorganisms (P<0.05).

The broad-spectrum antibacterial property even in antibiotic resistant and nosocomial pathogens of Surgi ORC is with reference to the intrinsic characteristics of oxidized cellulose, due to its low pH (3.0 to 4.0). Since low pH affects a relatively broad spectrum of bacteria and does not act in a mechanism-specific manner, as do antibiotics.

Br J Phar Med Res Copyright©2021 Dr Vidya Sagar et al. This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 International License (http://creativecommons.org/licenses/by/4.0/), allowing third parties to copy and redistribute the material in any medium or format and to remix, transform, and build upon the material for any purpose, even commercially, provided the original work is properly cited and states its license.

Keywords:
Antibacterial, ORC, Antibiotic resistance, Nosocomial

Corresponding Author: Dr Vidya Sagar, Head – R&D / QA & RA, Aegis Lifesciences Pvt Ltd, Ahmedabad, Gujarat, India – 382213.
INTRODUCTION:
The continued sustained progress of antibiotic-resistant microorganisms creates a critical problem for patients undergoing surgical procedures. In the event of an infection caused by an antibiotic resistant microorganism, treatment, and recovery will become problematic. It is therefore prudent to utilize products in surgery that minimize the risk of infection. Prevention of surgical site infections (SSIs) is of primary importance for the patient before it becomes problematic.

Secondly the nosocomial infections also called “healthcare associated infections” referred for the infections caused by hospital stay and it accounts for a major risk factor for serious health issues leading to death [1]. About 75% of the burden of these infections is present in developing countries [2,3]. Asymptomatic patients may be considered infected if these pathogens are found in the body fluids or at a sterile body site, post operation, such as blood or cerebrospinal fluid [4]. Infections that are acquired through hospital staff, visitors or other healthcare personnel may also be considered as nosocomial infections. Studies conducted in different parts of the world show that in North America and Europe 5%–10% of all hospitalizations result in nosocomial infections, while Latin America, Sub-Saharan Africa and Asia show more than 40% hospitalizations with nosocomial infections [5]. Nosocomial infections can be caused by any organisms, but few are particularly responsible for hospital-acquired infections at post-operative surgical sites.

National Healthcare Safety Network with Centre for Disease Control (CDC) for surveillance has classified nosocomial infection sites into 13 types, with 50 infection sites, which are specific based on biological and clinical criteria. The sites which are common include Urinary Tract Infections (UTI), surgical and soft tissue infections, gastroenteritis, meningitis, and respiratory infections. A change regarding nosocomial infection sites can be easily detected with time due to the elevated use of cancer chemotherapy, advancement in organ transplantation, immunotherapy, and invasive techniques for diagnostic and therapeutic purposes [6].

Oxidized Regenerated Cellulose (ORC), developed over 60 years ago, is a plant derived, passive Hemostat that promotes hemostasis by providing a matrix for platelet adhesion and aggregation. It is available in various fabric formats allowing for multiple applications, can conform to irregular surfaces and hard-to-reach sites, adheres easily to bleeding surfaces, and is suitable for many surgical procedures [7]. ORC is rapidly absorbed by hydrolysis within 7–20 days depending on the amount used. [8,9], ORC has bactericidal properties attributable to its local pH-lowering effect against a wide spectrum of pathogens [10,11]. Oxidized regenerated cellulose (ORC) is used broadly in surgical procedures as a hemostatic agent. Oxidized regenerated cellulose is produced by the oxidation of cellulose with nitrogen tetroxide [12]. ORC has been evaluated in prospective randomized studies, despite its widespread use in surgical practice [13,14], hence evidence-based information on its optimal use is limited.

A general discussion of the in vitro and in vivo antimicrobial properties of ORC was first reported by Dineen [10]. In vitro studies indicated that ORC had antimicrobial activity against a broad range of pathogens. These studies utilized zone of inhibition assays, as well as quantitative recovery of a broad range of challenge organisms using ORC. In an infected wound model in guinea pigs, ORC demonstrated in vivo activity in the prevention of sepsis as compared to control animals [10]. In an in vivo infected splenectomy model study in mongrel dogs, Dineen demonstrated that ORC was effective in reducing a bacterial population following a challenge [15]. In another mongrel dog study, wrapping an aortic patch with ORC prior to bacterial challenge resulted in a reduction of bacterial contamination.

Of the Several ORC hemostats available in the market, Surgi ORC is one brand of ORC product manufactured and marked by Aegis Life Sciences Pvt Ltd, India. It is used to assist control of bleeding from capillary, venous and small arteriolar vessels. In emergency/trauma situation Surgi-ORC can assist well in Haemostasis and serve as a haemostatic adjunct in the control of local haemorrhage. Surgi
ORC is available in different variants for different clinical applications during surgeries. The knitted variants of Surgi–ORC, ORIGINAL and KNIT are indicated for use in Cardiovascular and Thoracic Surgeries, General Surgeries including Cholecystectomy, Liver and Spleen Laceration, Port Site Bleeding, Tonsillectomy and Skin Grafting. The unknit variants of Surgi–ORC, FIBRIL and NON-WOVEN or SNOW models are mainly intended for use in different surgeries including Neurosurgery Laminectomy, Surgery on Tumour Bed, Head Trauma, Spine and Spinal Cord. It is so versatile because of its structure and design it can also be used in Cardiovascular and Thoracic Surgeries of Carotid Endarterectomy, Abdominal Aortic Aneurysm, Sternum Closure in CABG and Valve Repair/Replacement and Orthopaedic, Gynaecological Surgeries, Laparoscopic hysterectomy, and other General surgeries. However, the use of the variants is to discretion of the physician and not of patient’s choice. The present study reports the invitro antibacterial activity of a commercial brand of ORC (Surgi–ORC, Aegis Lifesciences Pvt Ltd, India) on antibiotic resistance and nosocomial pathogenic strains.

MATERIALS AND METHODS

Thirty-one strains of microorganisms of ATCC strains (some are clinical isolates) were studied by Microbial challenge test on recovery plates on four variants of Surgi–ORC. Inoculum of bacterial test strains were prepared by inoculating broth medium with stock cultures of test organism and incubating at 30–35°C for 24 h. All test strains were grown in Tryptone soya broth (TSB, Hi Media laboratories, Mumbai India) except for some organisms which were grown in the respective media conducive for their growth and temperature. This study was designed to evaluate the effect of Surgi ORC product variants against the microbial challenge over a period of 24 h. Four different Surgi ORC variants (ORIGINAL, KNIT, FIBRIL and NON-WOVEN / SNOW) were evaluated in all thirty-one microorganisms.

The products are referred to as ORC-O, ORC-K, ORC-F and ORC-SW respectively in this study. Each ORC sample was aseptically cut and weighed to 215 mg. Samples were placed into a sterile test tube and inoculated with 0.1 ml of the challenge microorganism from 24hr culture. A broth volume of 11 ml was added to each test tube, providing an approximate ratio of 20 mg ORC/ml media. This weight/volume ratio was based primarily on previous studies by Dineen [10], where 3 cm² of ORC was added to 10 mL of culture broth to assess antimicrobial activity. The assay medium for all test strains was TSB, with some exceptions. Following addition of broth medium, each test tube was vortexed for 2 minutes. Aliquots were removed from the prepared test tubes and were subsequently diluted with 0.85% sterile saline and pour plated. TSA was used for recovery plating of all challenge microorganisms except for some organisms. This procedure was carried out in 6 plates at 24 hr after inoculation. Positive controls of all test strains were
run alongside ORC challenge tests, and these were plated at time 0 and 24 h. All recovery plates were incubated at 30–35°C for 24 hr. Plates were read subsequently and results were recorded as colony forming units (CFU)/ml. The results were calculated as Mean SD of the CFU of the triplicates for each organism / variant and statistically compared with triplicate data of the positive controls samples. Statistical analysis was performed by ANOVA using Graph Pad Prism 7, Graph Pad Software, Inc., La Jolla, CA, USA

RESULTS
The results were depicted as Mean± SD of CFU and P value for each Surgi – ORC variant for all strains of organisms compared to positive controls in Table 1 and 2. The statistical significance compared to positive control for each Surgi ORC variant to corresponding organisms are provided in the respective tables.

Table 1: CFU values (Mean ± SD, n=6) of Surgi ORC variants against antibiotic resistant microbial strains and statistical significance (P value)

<table>
<thead>
<tr>
<th>Bacterial strain / ATCC no.</th>
<th>Control</th>
<th>ORC-O</th>
<th>ORC-K</th>
<th>ORC-F</th>
<th>ORC-SW</th>
</tr>
</thead>
<tbody>
<tr>
<td>MRSE (ATCC 51625)</td>
<td>23.6 ±7.3</td>
<td>4.8 ±1.9***</td>
<td>6.5 ±1.8***</td>
<td>6.4 ±1.4***</td>
<td>4.6 ±1.9***</td>
</tr>
<tr>
<td>MRSA (ATCC 33591)</td>
<td>25.8 ±6.6</td>
<td>5.1 ±1.6***</td>
<td>3.6 ±1.2***</td>
<td>4.7 ±1.1***</td>
<td>4.2 ±1.2***</td>
</tr>
<tr>
<td>VRE (ATCC 51299)</td>
<td>20.6 ±8.4</td>
<td>8.2 ±1.9***</td>
<td>7.8 ±2.1***</td>
<td>7.0 ±2.4***</td>
<td>6.1 ±2.8***</td>
</tr>
<tr>
<td>PRSP (ATCC 49619)</td>
<td>18.6 ±5.2</td>
<td>4.7 ±1.8***</td>
<td>3.3 ±1.1***</td>
<td>3.0 ±1.7***</td>
<td>4.9 ±1.2***</td>
</tr>
<tr>
<td>MDRPA (ATCC BAA-2108)</td>
<td>21.3 ±6.5</td>
<td>14.6 ±2.4**</td>
<td>15.3 ±2.8**</td>
<td>14.9 ±3.1**</td>
<td>16.1 ±2.4**</td>
</tr>
<tr>
<td>MDRKP ATCC (BAA-1705)</td>
<td>30.7 ±7.5</td>
<td>19.8 ±1.5**</td>
<td>19.1 ±2.9**</td>
<td>21.3 ±3.1**</td>
<td>22.1 ±2.9**</td>
</tr>
<tr>
<td>MDR CD (ATCC 43255)</td>
<td>24.4± 6.9</td>
<td>18.8 ±5.7*</td>
<td>19.0 ±8.4*</td>
<td>20.4± 7.9*</td>
<td>15.6± 8.1*</td>
</tr>
</tbody>
</table>

*P<0.05, **P=0.01, ***P<0.001


Table 2: CFU values (Mean ± SD, n=6) of Surgi ORC variants against non-antibiotic resistant and other pathogenic microbial strains with statistical significance (P value)

<table>
<thead>
<tr>
<th>Bacterial strain / ATCC no.</th>
<th>Control</th>
<th>ORC-O</th>
<th>ORC-K</th>
<th>ORC-F</th>
<th>ORC-NW</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pseudomonas aeruginosa (ATCC 9027)</td>
<td>28.6 ±9.5</td>
<td>4.7 ±1.4***</td>
<td>6.7 ±2.1***</td>
<td>4.5 ±2.0***</td>
<td>3.8 ±1.5***</td>
</tr>
<tr>
<td>Streptococcus haemophilus (ATCC 27734)</td>
<td>34.5±10.9</td>
<td>24.7±15.4*</td>
<td>22.3±15.4*</td>
<td>27.7±14.6*</td>
<td>20.4±13.9*</td>
</tr>
<tr>
<td>Staphylococcus aureus (ATCC 6538)</td>
<td>32.3±11.2</td>
<td>7.6 ±2.1***</td>
<td>8.5 ±2.4***</td>
<td>4.8 ±2.9***</td>
<td>4.2 ±2.4***</td>
</tr>
<tr>
<td>Bacillus cereus (ATCC 14579)</td>
<td>30.7±9.8</td>
<td>10.8 ±2.6***</td>
<td>8.3 ±2.7***</td>
<td>12.8±2.1***</td>
<td>10.0 ±3.9***</td>
</tr>
<tr>
<td>Streptococcus pyogenes (ATCC 12344)</td>
<td>24.1±4.6</td>
<td>11.4 ±3.4***</td>
<td>10.2 ±3.2***</td>
<td>14.2±1.2***</td>
<td>9.6±2.4***</td>
</tr>
<tr>
<td>Micrococcus luteus (ATCC 9341)</td>
<td>24.9±4.4</td>
<td>16.9 ±2.6**</td>
<td>20.4±2.5**</td>
<td>19.4±2.8**</td>
<td>18.6±2.7**</td>
</tr>
<tr>
<td>Streptococcus salivarius (ATCC 13419)</td>
<td>28.2±3.4</td>
<td>10.8 ±2.8***</td>
<td>12.4 ±2.8***</td>
<td>13.8±2.4***</td>
<td>10.8±4.4***</td>
</tr>
<tr>
<td>Moraxella catarrhalis (ATCC 25238)</td>
<td>27.4±7.4</td>
<td>18.7 ±3.9**</td>
<td>21.4±1.4**</td>
<td>19.1±1.7**</td>
<td>19.9±3.7**</td>
</tr>
<tr>
<td>Klebsiella aerogenes (ATCC 13058)</td>
<td>32.8±5.8</td>
<td>13.4 ±3.2***</td>
<td>14.1±3.9***</td>
<td>13.2±2.4***</td>
<td>10.1±2.8***</td>
</tr>
<tr>
<td>Shigella dysenteriae (ATCC 13048)</td>
<td>29.5±5.4</td>
<td>19.9 ±2.9**</td>
<td>23.1±3.4**</td>
<td>18.7±3.7**</td>
<td>18.7±2.7**</td>
</tr>
</tbody>
</table>
Table 1 provides the mean ± SD CFU values of each strain of microorganism for the respective Surgi ORC variant. The data also provides the level of significance (P value) compared to their positive control values.

It can be categorically understood from the CFU values and statistical analysis that the four Surgi ORC variants are having extremely significant (P<0.001) antibacterial activity on - Methicillin Resistant Staphylococcus epidermidis (MRSE), Methicillin Resistant Staphylococcus aureus (MRSA), Vancomycin-Resistant Enterococcus faecalis (VRE) and Penicillin-Resistant Streptococcus pneumoniae (PRS). The two Multi Drug Resistant (MDR) strains of organisms, Klebsiella pneumonia and Pseudomonas aeruginosa has moderate but very significant (P<0.01) effect with all four Surgi ORC variants compared with the respective positive control values. However, all the four Surgi ORC variants have shown significant (P< 0.05) but mild antibacterial activity on MDR Clostridium difficile.

The Surgi ORC variants have shown significant and considerable antibacterial activity on other organisms which are not resistant to antibiotics but are potential nosocomial pathogens at various post-operative surgical sites.

Table 2 provides the mean ± SD CFU values non antibiotic and other pathogenic strain of microorganism for the respective Surgi ORC variants. The data also provides the level of significance (P value) compared to their positive control values. There were large differences with CFU values of the positive controls of each microbial strain (18 to 38 CFU).

Of the 24 microbial strains tested in this category, Surgi ORC showed extremely significant microbial activity in 13 strains (P<0.001), Very significant and moderate activity in 7 strains (P<0.01) and mild activity in 4 strains (P<0.05). Microbes of Enterococcus and Staphylococcus family presented very high antimicrobial activity with all four variants of Surgi ORC. Those strains showed 70% to 90% inhibition compared to positive controls. Serratia marcescens, Mycobacterium phlei, Corynebacterium xerosis, Streptococcus haemophilus exhibited mild antimicrobial activity amongst the 24 microbial strains.

**DISCUSSION**

Oxidized regenerated cellulose (ORC) is used broadly in surgical procedures as a hemostatic agent. Whereas there are several products on the market that also are used for hemostatic applications, ORC is the only such
agent with known broad-spectrum antimicrobial activity. The antibacterial property ORC Haemostats is with reference to the inbuilt characteristics of Oxidized Cellulose itself. The cause of the antibacterial effect is due to the low pH (3.0 to 4.0) of Oxidized Cellulose, which inhibits the growth of microorganisms. ORC is also effective against antibiotic resistant organisms that too attributed to its very low pH. The pH of 4.4 to 9 is the limiting range for many organisms \[16\], including staphylococci, Pseudomonas, streptococci, coliforms, and others commonly associated with medical device–related SSIs. Since low pH affects a relatively broad spectrum of bacteria and does not act in a mechanism-specific manner, as do antibiotics. Apart from all the spectra of microbes tested, Gram-positive bacteria are of particular concern, as they comprise the three most common pathogens isolated from SSIs and are frequently antibiotic-resistant. These include Staphylococcus aureus, coagulase-negative staphylococci, and Enterococcus species \[17\]. Nosocomial pathogens which also include some antibiotic resistance organisms pose a serious threat to infections at surgical sites post operatively. Surgi – ORC has shown extremely significant antibacterial activity against all the related organisms.

In the present study, it is noted that ORC-F and ORC SW more effective against in general on all strains of microorganisms than ORC-O and ORC-K. CFU values at 24-h exposure show an average of 15 % more antibacterial activity of ORC -F and ORC - SW. It may be due to the physical configuration of the product matrix or product knitting pattern. A very loose, relatively thick matrix with a cotton–like appearance will provide more surface and contact area. Hence there is advantage of using Surgi ORC as haemostat, which has broad spectrum antibacterial activity in controlling bleeding during and post-surgery. It is in additional to the general, hospital manual directed, antibiotic regimen during surgeries. However, it is advised that Surgi ORC, cannot be an alternative to systemically applied therapeutic or prophylactic antimicrobial agents.

CONCLUSION

From the studies its evident that Surgi ORC is having broad spectrum antibacterial activity, even against antibiotic resistant and nosocomial pathogens. It is attributed to its low pH, an intrinsic characteristic of the material and not of any medicinal component. Hence it is advantageous of using Surgi ORC as haemostat with additional claim of antibacterial activity in controlling bleeding during and post-surgery.

REFERENCES


How To Cite This Article:

Source of Support: Nil
Conflict of Interest: None declared

Your next submission with British BioMedicine Institute will reach you the below assets
• Quality Editorial service
• Swift Peer Review
• E-prints Service
• Manuscript Podcast for convenient understanding
• Global attainment for your research
• Manuscript accessibility in different formats (Pdf, E-pub, Full Text)
• Unceasing customer service

Track the below URL for one-step submission
https://www.bjpmr.org/manuscript-submission/