含銀活性碳敷料應用於抗菌效果與傷口癒合評估

Influence of Various Ag-containing Wound Dressing in Wound Healing

Wan-Yu Chung[1], Wei-Shan Hsu[1], Yu-Hsin Lin[2]* and Jui-Hsiang Lin[1]*

[1] 科云生醫科技股份有限公司 Bio-Medical Carbon Technology Co., Ltd. Taichung, Taiwan

[2] 中國醫藥大學生物科技系 Department of Biological Science and Technology, China Medical University

*Corresponding Email: ylhsin@mail.cmu.edu.tw; goni.lin@bm-carbon.com

摘要

銀離子具有廣效性抑菌效果，含銀活性碳纖維廣泛應用於傷口癒合和傷口感染治療，本研究室和科云生醫科技股份有限公司共同探討不同類型含銀的敷料對傷口修復之影響。實驗結果得知，不同銀含量的敷料其皆具有銀離子釋放特性並達極佳抑菌能力。含銀活性碳纖維在細胞測試得知仍具有良好生物相容性。動物實驗的大鼠傷口癒合得知，市售的含銀敷料和科云生醫科技股份有限公司所研發的含銀活性碳纖維，皆達到膠原蛋白新生和傷口組織再生特性。

Abstract

Silver has been applied on wound care because of its wide-spectrum antimicrobial activity and rare incidence of resistant bacteria developed. Silver containing dressings have been widely used for controlling wound infection; however, the association of its antimicrobial activity and wound healing improvement with the silver concentration remains unclear. We studies various Ag-containing activated carbon fiber (in cooperation of Bio-Medical Carbon Technology) for understanding the influence of the silver concentration on efficacy of an Ag-containing dressing. Various Ag-containing activated carbon fiber exhibited high biocompatibility; silver concentration showed a minor influence on cytotoxicity. The infected excisional wound model in rat indicated that compared to other commercial Ag-containing dressings, Ag-containing activated carbon fiber assisted wound healing in early phase by promoting granulation and collagen deposition.

Introduction

Antibiotics are once the powerful medicines for controlling pathogenic infection. However, the emerge of various antibiotic-resistant bacteria like Methicillin-resistant *Staphylococcus aureus*, *Pseudomonas Aeruginosa* and *Acinetobacter baumannii* impedes and limits the broad efficacy of antibiotics [1,2]. The antimicrobial activity of silver results in change of bacterial membrane permeability, inhibition of bacterial DNA replication/RNA transcription and dysfunction of bacterial enzymes [3,4]. The safety and effectiveness of silver-containing dressings have been widely accepted and been verified by plenty of clinical trials [4]. Activated carbon has been applied on wound dressing which is originally designed for controlling wound malodor [4]. A commercial wound dressing composed of polyacrylonitrile (PAN)-based activated carbon fiber supporting with silver particle by impregnated method, KoCarbonAg®, has been developed by Bio-Medical Carbon Technology Co., Ltd. In this study, we aim to understand how silver content on PAN-based activated carbon fiber associates with its antimicrobial activity and wound healing, at the meantime, using the commercially available Ag-containing dressings above as the comparison.

Materials and method

Preparation of ACF and various Ag-containing ACF and silver release

The raw activated carbon fiber (ACF) cloths in this study came from Bio-Medical Carbon Tech., Taiwan. The samples were then immersed in 3 ml distilled water, then subjected to inductively coupled plasma-optical emission spectrometer (ICP-OES) to determine the silver concentration.

Antimicrobial activity

Bacterial suspension at indicated time intervals were serial diluted and plated on the TSB agar plates. The colony-forming unit of bacterial suspension exposed to dressing was determined by counting the amount of colonies.

Animal study of wound healing and distribution of Ag of dressings on intact skin

The wounded rats each received 100 μL of the bacterial mixture including *P. aeruginosa*, *E. coli* and *S. aureu* on the wound surface. The wounds were photographed and examined to determine wound size reduction. And, the prepared samples (blood) were subjected to ICP-OES to determine the silver content (ppm) of the test sample.

Results and discussion

Micrographs of Ag-containing ACF wound dressings

ACF containing silver with 1 mg, 5 mg, 10 mg and 20 mg per 100 cm^2 (Ag-1/ACF, Ag-5/ACF, Ag-10/ACF and Ag-20/ACF) were prepared and investigated their morphology under a scanning electron microscope. The silver particles with 30 nm in diameter (black arrows) are distributed along the fiber and quantity is in proportion to Ag content indicated. The KoCarbonAg®, contains silver with an average of 5 mg per 100 cm^2, which is represented as Ag-5/ACF (Fig. 1).

***In vitro* antimicrobial activity**

The result mentioned above, even though these various Ag-containing dressings (Ag-1/ACF, Ag-5/ACF, Ag-10/ACF and Ag-20/ACF) exhibited different efficiency for bacterial elimination during the first couple hours, they all showed approximately 90% inhibition effect at 24 hours for all three bacterial strains that were tested (Fig. 2).

Effect of various Ag containing ACF on healing of infection wounds

The combination of *P. aeruginosa, S. aureus* and *E. coli* were used to introduce infection on the excisional wound. In general, Ag-20/ACF exhibited the greatest reduction of wound area in this study. This result indicates that Ag-20/ACF and Ag-5/ACF accelerate the early phase of infected wound healing; presumably due to its high biocompatibility to wound tissue as well as its antimicrobial activity to control infection, leading to the shorter inflammatory phase and the early entry of proliferation phase. The result indicates that once the infection is under control, the healing rate is then less affected by the silver concentration of Ag-containing ACF (Fig. 3).

Histological evaluation of infected wound healing

The histological results for hemaoxylin-eosin stain of infected wound treated with Ag-5/ACF and Actisorb silver 220. Both Ag-5/ACF and Actisorb Silver 220-treated infected wounds presented aggregations of neutrophils and macrophages as the inflammatory response occurs. Ag-5/ACF-treated infected wound exhibited certain degree of granulation (Fig. 4, yellow arrow). Actisorb Silver 220-treated infected wounds showed less granulation tissue formation but more neo-capillary formation than Ag-5/ACF (Fig. 4, black arrow).

Distribution of silver

The Ag-containing dressings were applied on wound area, serum silver level increases during the applied period, however, the level decreases to less than 0.1 ppm 21 days after removal of the dressing (Fig. 5), which is within the normal range of silver concentration in mammalian tissue.

Conclusion

The ACF with various Ag concentrations tested in this study, released silver ion in proportion to its silver content. *In vivo* study indicated that various Ag-ACF can promote infected wound healing in early phase compared to gauze and two other commercial Ag-containing dressing. Histological results indicate Ag-ACF promotes granulation and collagen deposition of the infected wound.

References

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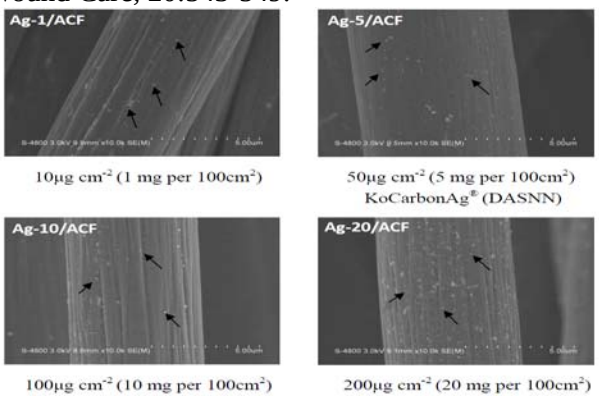

Fig. 1. Micrographs of distinct Ag/ACF dressing.

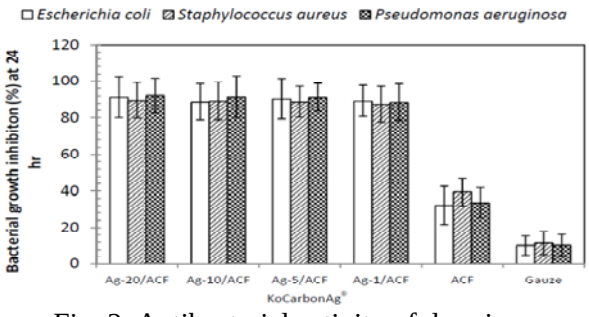

Fig. 2. Antibacterial activity of dressings

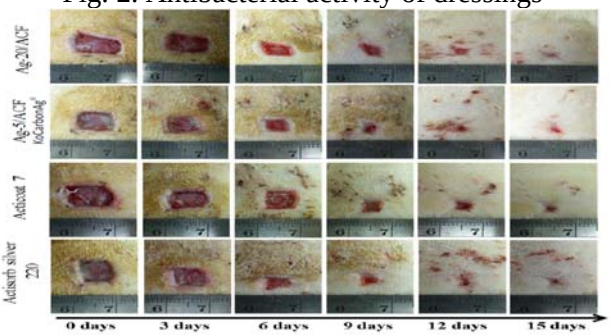

Fig. 3. Representative photographs of the infected wound with treatment of Ag-containing dressings.

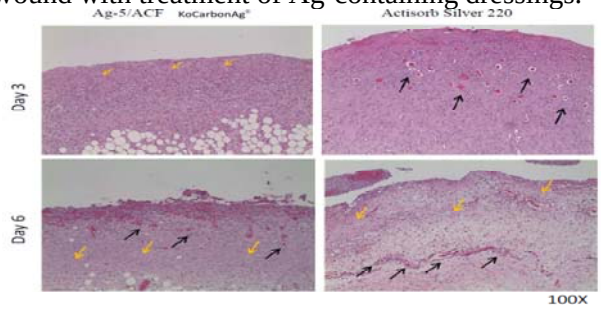

Fig. 4. Histologic image of the infected wound treated with Ag-5/ACF and Actisorb Silver 220

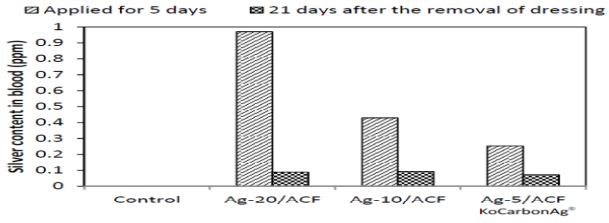

Fig. 5. Serum silver level after applying dressing