International Journal of Biological Macromolecules 117 (2018) 25–29

Contents lists available at ScienceDirect

International Journal of Biological Macromolecules

journal homepage: http://www.elsevier.com/locate/ijbiomac

Skin protectant textiles loaded with fish collagen, chitosan and oak galls extract composite

Ahmed A. Tayel [a,*], Reem A. Ghanem [a], Shaaban H. Moussa [b], Mona Fahmi [a], Hajar M. Tarjam [a], Nadia Ismail [a]

[a] Faculty of Aquatic and Fisheries Sciences, Kafrelsheikh University, El-Geish St., 33516 Kafrelsheikh City, Egypt
[b] Genetic Engineering and Biotechnology Research Institute, University of Sadat City, Egypt

ARTICLE INFO

Article history:
Received 10 April 2018
Received in revised form 19 May 2018
Accepted 21 May 2018
Available online 22 May 2018

Keywords:
Antibiotic-resistant
Antimicrobial
Dermal disinfection
Durable fabrics

ABSTRACT

Skin protection and control of its microbial pathogens are highly important demands; natural biological agents are the ideals for that. Collagen (Cg) was extracted and characterized from skin and scales of Nile tilapia fish (*Oreochromis niloticus*), chitosan (Cts) was extracted from shrimp shells and extract of oak (*Quercus infectoria*) galls (OGE) was prepared. The antimicrobial potentialities of extracted agents, Cts and OGE, were qualitatively proved against skin pathogens, *Staphylococcus aureus* and *Candida albicans*, including both antibiotic sensitive and resistant strains, neither Cg nor negative control exhibited antimicrobial actions toward examined strain. The entire agents were loaded onto cotton fabrics and evaluated for antimicrobial actions and durability. Loaded textiles with the combined extracts' composite were the most effectual followed by individual treatments with OGE and Cts, respectively. Treated textiles upheld most of their antimicrobial activity after 2 laundering cycles toward all microbial pathogens. This invention could be consequently applied for production of skin protectant and hygienic fabrics.

1. Introduction

Skin is the first human defense barrier that has many vital roles in body protection from external invasions and infections; severe skin impairments would unfaithfully affect the patient look, lead to disastrous infections, loss of water and electrolyte disorders and even could cause death [1]. The key challenge for skin caregivers is the emergence of infection from antibiotic-resistant microorganisms in traumatic wounds [2]. Although many broad spectrum antibiotics were implicated for control of these resistant strains, the antibiotic prophylaxis is regularly not sufficient to reduce wound bioburden bacteria under combat conditions [3]. For potential overcoming the resistance problem, novel antibiotics alternatives should be screened to treat wound infection.

Collagen (Cg) is from the major extracellular protein matrix in dermal tissues, which plays a dynamic role in mediating cell migration and adhesion because of its specialized recognition interaction. Cg-based dermal formulations were faithfully applied for skin regeneration, with designating low immunogenicity, high biocompatibility and high porosity [4].

Cg could be extracted from many living organisms, e.g. cows, pigs, fish …; the Cg produced from aquatic organisms is closing to human Cg, could convince religious requests like halal (Islam) and kosher (Judaism), and also augment economic impact from fishery industry [5]. The resulted wastes/by-products from fisheries processing industries, e.g. fish skins and bones, shrimp shells …, could be over 75% of the catch total weight [5]. Many Cg-based products were lately developed, approved and commercialized for application in wound healing [6].

Chitosan (Cts) is a derived biopolymer from chitin, after deacetylation, which have numerous applications in environmental, nutritional, biotechnological and health care fields [7]. The Cts antimicrobial and biomedical characteristics, including regenerative medicine, were discussed and confirmed from many studies [8,9].

Due to its biocompatibility, antimicrobial and intrinsic hemostatic properties, Cts was recommended as an attractive biomaterial for inclusion in wound care dressings [10].

Cts was recurrently applied, as a skin substitute biomaterial, in dermal tissue engineering because of its haemostasis properties, which support tissues regeneration and motivate the collagen fibroblasts synthesis [11,12].

Oak (*Quercus infectoria*) galls are the arising abnormal growth on the young twigs of trees after invasion by oak swap [13]. The oak galls extract (OGE) have elevated contents from tannins; the main identified OGE constituents were gallotannic, gallic and ellagic acids, along with diverse carbohydrates [14]. OGE was applied, through folkloric medicine, to prevent and treat several diseases and disorders, and proved to have potent antimicrobial, antioxidant, larvicida, anti-inflammatory

* Corresponding author.
E-mail addresses: tayel_ahmad@yahoo.com, ahmed_tayel@fsh.kfs.edu.eg (A.A. Tayel).

https://doi.org/10.1016/j.ijbiomac.2018.05.150
0141-8130/

and antiviral actions [13,15,16]; it was also recommended for its wound healing potentiality [17,18].

The "smart" fabrics was suggested to overcome the potential dermal infections/harms; the main bases for biomaterials applications, e.g. Cg and Cts, as wound dressings are their ideal characteristics to supply the skin interface with moist environment, function as microbial barriers, with gaseous exchange allowance and excess exudates removal. They should have also a non-allergenic, non- toxic and non-adherent attributes, and preferably made from a minimally-processed biomaterials that have wound healing and antimicrobial potentialities [19].

Therefore, current investigation aimed to produce Cg, Cts and OGE and evaluate their antimicrobial potency against skin pathogens, including antibiotic resistant types, and to utilize them for developing antimicrobial and skin protectant textiles with favorable characteristics.

2. Materials and methods

2.1. Fish collagen extraction and characterization

Nile tilapia (*Oreochromis niloticus*) filleting by-products (e.g. fish skins and scales) were obtained from the Fish Processing Research Plant, Kafrelsheikh University. The collected samples were kept at −20 °C until using then thawed and subjecting to extraction process. Minor modifications from the described of Nagai and Suzuki [20] were applied; the entire steps were conducted at ≤4 °C, using double distilled water (DW) for dissolving and washing. Briefly, skin was cut into ~2 cm^2 pieces then samples, skin or scales, were washed and non-collagenous proteins were deproteinized using 0.1 N NaOH, rewashed and lyophilized. Collagen was extracted twice using immersion in 0.5 M acetic acid for 48 h, filtration and salting out with 1.0 M of NaCl. The precipitated acid soluble collagen (Cg) was collected by centrifugation at 20.000 ×*g* for 45 min, then the pellets were dissolved acetic acid (0.5 M) and dialyzed in diluted acetic acid (0.1 M) and DW, collected and lyophilized. The Cg were characterized by SDS-polyacrylamide based gel electrophoresis [21], and by spectroscopy (Fourier transform infrared - FTIR) [22].

2.2. Shrimp shell chitosan preparation

Chitosan (Cts) was extracted from shrimp (*Penaeus monodon*) exoskeletons wastes, obtained from the Aquaculture Research Farm – Kafrelsheikh University – Egypt, as demonstrated by Lamarque et al. [23]; the shrimp flesh were removed from exoskeletons, which were then repeatedly washed with DW, dried at 50 °C for 24 h and crushed with mechanical grinder. The deproteinization step was conducted using hot 0.1 M NaOH solution (~75 °C) for 2 h, then cooled, filtered, recurrently washed to eliminate NaOH residues, dried and grinded again to fine powder. Deproteinized shells were then treated with 0.2 M of HCl for 24 h at 24 °C for the removal contained minerals (demineralization), then the HCl was drained and resultant chitin was continually washed. The deacetylation step was performed in chitin using solution of 60% NaOH and heating at 121 °C for 90 min; after cooling, the formed chitosan was filter separated, extensively washed and dried at 65 °C for 12 h.

The chitosan was subjected to FTIR analysis to calculate its deacetylation degree [22], whereas the Cts molecular weight was determined using liquid chromatography with high-performance (HPLC), according to Wu et al. [24].

2.3. Oak galls extraction

Dried galls of *Q. infectoria* were obtained from ARC, Giza, Egypt. With an electric grinder, the crushed galls were powdered to acquire particles with ~70-mesh size. The galls powder (250 g) was immersed in 1250 mL of 70% ethanol and shaken for 350 min in a rotary shaker (at 120 ×*g*). Galls extracts (OGE) were then filtered, in a Buchner funnel, through filter paper and pooled for flash evaporation (Büchi, Flavil, Switzerland) at reduced pressure at 45 °C, to omit most of solvent contents and then desiccated under vacuum for 6 h. The resulted dry OGE weight was recorded and resuspended in sterile tween 80 aquas solution for usage in further experiments.

2.4. Microbial skin pathogens

Different skin microbial pathogens were used for evaluation of the products antimicrobial activity. The used strains were *Candida albicans*-S (ATCC-10231), *C. albicans*-R (resistant strain to fluconazole, isolated from human skin lesion), *Staphylococcus aureus*-S (ATCC-25923) and *S. aureus*-R (methicillin resistant strain, isolated from infected wound). The microbial strains were propagated and screened at 37 °C using yeast/malt extract agar, for *C. albicans* strains, and trypticase soy agar, for *S. aureus* strains.

2.5. Antimicrobial assays

The growth inhibition zones (IZ) appearance, after well diffusion assay, was measured as indicator for antimicrobial activity from Cg, Cts and OGE [25] Diluted solutions from the extracted compounds, with concentration of 1% (w/v), were made in 1% acetic acid solution. 100 μL from each microbial cell suspension (~2×10^6 CFU) were spread onto appropriate agar media, then wells of 6 mm were made in agar and impregnated with 50 μL from diluted extracts. Plates (in triplicates) were incubated at 37 °C for 18 h, and appeared IZs diameters' means were calculated. Diluted acetic acid (1%) was used as negative control.

2.6. Antimicrobial textiles preparation

Scoured cotton plain weave (108 g/m^2), from Misr Weaving and Spinning Co - Egypt, were sterilized and treated with the skin protectant

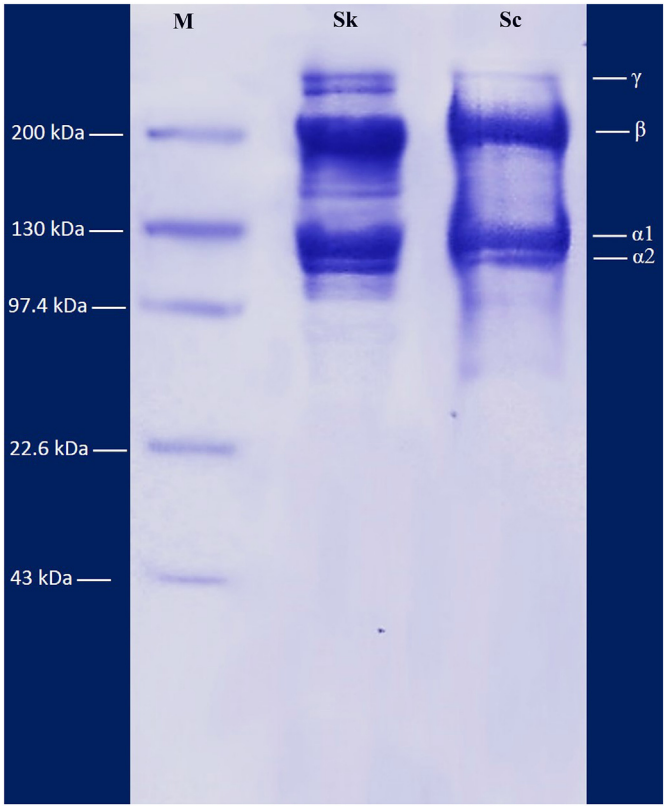

Fig. 1. SDS–PAGE pattern of produced collagen from fish skin (Sk) and scales (Sc) compared to protein marker (M).