REVIEW ARTICLE

Larval Therapy as a Wound Healing Acceleration in Chronic Wounds : A Scoping Review

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ABSTRACT

The success of the wound healing process depends on effective debridement and infection control. Currently, some difficulties are encountered in controlling infected chronic wounds due to the development of resistance to antibiotics and the inability of surgical debridement of necrotic scar tissue. Larval therapy is capable of causing disruption of the biofilm, making it beneficial for chronic wound healing. The aim of this scoping review was to reveal the use of larvae for chronic wound healing, by inhibiting the formation of biofilms based on a number of articles that have been published in journals. Articles were searched using databases such as Pubmed, Google Scholar and Cochrane Library. We found articles related to the use of larval therapy as many as 15 articles between 2012-2020 that were eligible for review. In conclusion, chronic wound therapy using larvae is easy to apply and is successful as a traditional and complementary medicine.

Keywords: Antimicrobial, Biofilms, Chronic wound, Larval therapy, Medicine

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INTRODUCTION

Wounds are defined as impaired skin or mucosal integrity as a result of trauma. This necrotic condition that occurs in the skin and mucous membranes can close over time by a special mechanism (1). This condition is usually called wound healing and consists of four physiological stages namely homeostasis, inflammation, proliferation, and maturation (2). For wounds that do not heal or that close late, removal of tissue debris for various reasons, removal of damaging products such as local infection and/or proteases from the wound bed stops healing (3).

The success of the wound healing process depends on effective debridement and infection control. Currently, some difficulties are encountered in controlling infected chronic wounds due to the development of resistance to antibiotics and the inability to debride the necrotic scar tissue (4). This kind of wound appears as a wound that greatly reduces the quality of human life and also fails to heal because it does not go through the normal stages of healing, thereby increasing the cost of treatment (5).

Increased knowledge and understanding of healing, prevalence, incidence and complications of chronic wounds are increasingly complex. The presence of biofilms, as well as complications caused by chronic wounds are also obstacles and triggers for failure of wound healing (6). Bacterial biofilms are a group of polymicrobial organisms (fungi, bacteria, etc.) packaged in a exopolymeric matrix and have a toleration for host defenses, antiseptics and antibiotics (7). It's estimated that there are an average of 6.3 bacterial species in chronic wounds, which are dominated by Pseudomonas aeruginosa and Staphylococcus aureus (8). Therefore, alternative therapies are needed that can suppress the formation of biofilms in chronic wounds. Eradication of biofilm-forming bacteria is very difficult, so the best option is to remove the infected area if possible (biofilm disruption) accompanied by antimicrobial therapy. There are several ways to disrupt biofilms, including: ultrasound waves, negative pressure therapy, and larval therapy (9). Apart from being an antimicrobial and disrupting biofilms, larval therapy has been known to have various other beneficial effects on chronic wound healing, including: inflammatory response, complement system, fibrinolysis, and angiogenesis (10). Although studies on the effects of larval excreta/secretions have been carried out quite a lot, not many studies have reported on components that have antimicrobial and anti-biofilm effects in vivo. The aim of this review was to reveal the use of larvae for chronic wound healing, by inhibiting the formation of biofilms based on a number

of articles that have been published in journals.

METHODS

Data sources and searches

This scoping review was performed according the PRISMA guidelines. The articles were searched conducted using databases such as PubMed, Google Scholar and Cochrane Library. The inclusion criteria were as follows: (a) studies about larval therapy (b) literature review, case report and systematic review. In the early stages of searching for journal articles, 44 articles were obtained from 2012 to 2020 using the keywords "larvae treatment", "larva", "maggot debridement", "wound healing", "chronic wounds", and "biofilm".

Study selection

Following the removal of duplicates, the titles and abstracts of the identified records were independently reviewed by two reviewers (FFB and HSB). Following the primary question, the full texts and abstracts were identified and reviewed. Disagreements were then decided through discussion with the third investigator (NFA and DSE).

RESULTS

Study characteristics

Fig. 1 presented the electronic search process. We initially identified 44 articles. Among these, 10 duplicate articles and screening based on duplication as much 34. After reviewing the titles and abstracts, irrelevant studies were excluded 11 articles. Five full text articles were excluded because not providing full accessed. Three studies were excluded because of a lack of relevant data. We then retained 15 articles for further analysis in describing the effect of effects on chronic wound healing, including: inflammatory response, complement system, fibrinolysis, and angiogenesis with larval excreta/secretions and stimulation of granulation tissue formation.

Wound healing

Wound is a condition of damage to the continuity of tissue, anatomical structure and function of normal skin or mucosa due to pathological processes originating from the internal or external environment and affecting certain organs. Treatment and management of wounds in this case is one of the factors that determine the final outcome of the wound healing process (11).

Wound healing is a very dynamic process and involves various interactions between extracellular matrix molecules, mediators, tissue cells including blood vessels, and leukocyte infiltration (12). The wound healing process consists of several phases that integrate and overlap, namely: hemostasis; inflammation; mesenchymal cell differentiation, proliferation, and migration to the wound area; adequate angiogenesis; and

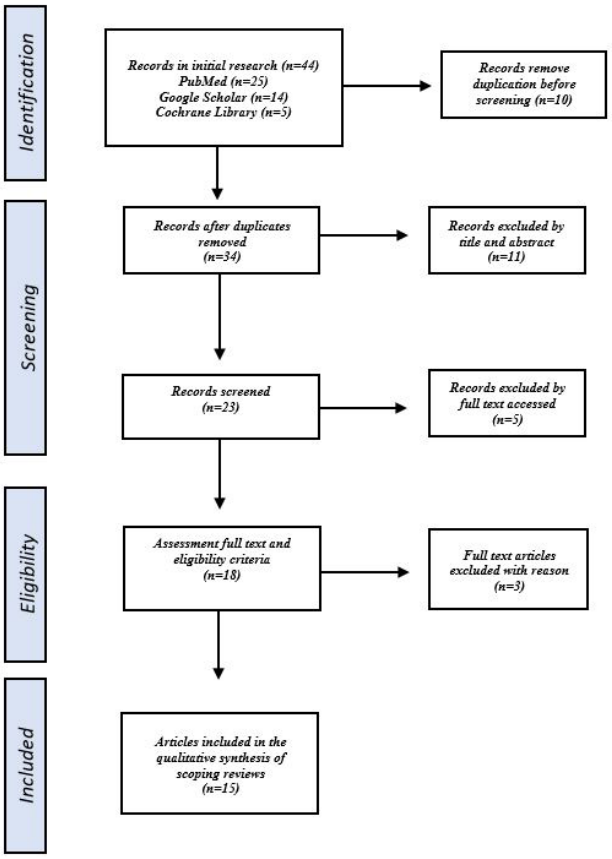

Fig. 1: PRISMA flowchart of the study selection process.

rapid re-epithelialization over the wound surface (13). Each phase must take place precisely and programmed. Any obstruction, deviation, or extension will lead to delayed wound healing or chronic nonhealing wounds (6).

The stages of wound healing are a complex process that occurs in stages consisting of stages of hemostasis, inflammation, proliferation and remodeling. At the beginning of the inflammatory phase, secreted chemotactic factors attract neutrophils and macrophages to destroy damaged tissue with the help of proteinases, Reactive Oxygen Species (ROS), and Reactive Nitrogen Species (RNS) (14). In the proliferative phase a number of growth factors will be secreted in large quantities so that it will trigger the release of Matrix Metalloproteinase (MMP) (15). MMP is a group of endopeptidases whose activity in large amounts is able to degrade the extracellular matrix. In addition, excessively secreted ROS and RNS can also cause toxic effects in the form of severe oxidative damage to the skin (11).

Wound healing is also influenced by factors in the body, namely IL-6, FGF-1, FGF-2, collagenase, H2O2, and BM-MSCs (16,17). Wound care can be done using microbial cellulose, wound dressings, or modified vacuum systems. The development of the formula of the system and the base used was also carried out to assist the wound healing process. Active substances from natural ingredients are also being intensively developed

as alternative treatments (11).

Biofilm

The biofilm is a population of microorganisms attached on a solid surface; where the surface is in the form of living tissue and dead tissue (4). Biofilm formation can occur on various types of surfaces and various environmental conditions where bacteria are present. Bacteria, organic and inorganic molecules that are on the surface then form a film condition (8). These organic and inorganic substrates together with microorganisms move to the surface by diffusion or following the flow of liquids. Nutrient transfer was higher in the biofilm than in the liquid phase (5). Biofilms are made up of bacteria and exopolysaccharides (EPS), which are self-produced extracellular polymeric molecules (18). The development of an intact biofilm containing multiple layers including an EPS matrix with a vertical structure, and film formation (19). The vertical structures of microorganisms are sometimes tower-like or mushroom-like, separated by interstitial spaces. Most of the biofilm can readily and quickly absorb nutrients from the surrounding fluid and eliminate by-products from the biofilm to the interstitial gap (20). Biofilm production is complicated, but it can be subdivided into four main steps: deposition and development of the biofilm, microbial (planktonic) adhesion to the film sheet, and bacterial growth and colonization (6).

Structure of Biofilm

The biofilms made up from microbial cells and extracellular polymeric material (EPS). EPS can cover between 50 and 90 percent of biofilm's organic total carbon and can be termed its main material matrix. Chemical and physical features of EPS might vary, however it is predominantly made of polysaccharides. Some polysaccharides, such as the EPS of gram-negative bacteria, are neutral or polyanionic. The existence of uronic acids, such as D-glucuronate, D-galacturonic, and mannuronic or pyruvic acid, induces the combined of divalent cations like as calcium and magnesium, that is demonstrated to cross-react with polymeric fibers that bring increased binding strength on biofilm development (21). Composition of EPS in gram-positive bacteria, like Staphylococci, can be highly varied and is predominantly cationic (22, 23). Although most EPS are hydrophilic and hydrophobic, their solubility varies (23). Important EPS features that may have a significant impact on biofilms. First, the polysaccharide content and structure establish the major confirmation of EPS, which is that many bacterial EPS contain a 1,3- or 1,4--hexose residue structure. They are more stiff, less deformable, and in certain circumstances insoluble, while the EPS molecule can dissolve easily in water. Second, EPS biofilms are generally uneven and can change location and are temporary (24).

When bacteria build biofilms, other bacteria are drawn to them by molecular pathways, resulting in a polymicrobial system that can survive. Long-lasting biofilms are frequently related with biofilm genetic diversity, leads to chronic wounds resistant to therapy. Gene expression is required for bacterial biofilm survival and host adhesion, host cell growth and development to prevent biofilm detachment, induce local inflammation, and stimulate plasma production in the wound layer to supply nutrients to the biofilm colony (25).

Larva therapy

The larvae has been used for medical applications in chronically infected wounds for centuries by the Maya Indians of Central America, the Australian Aborigines, and in China. In the past, larvae therapy was used to treat wound infections (Table). During World War I, Baer an orthopedic surgeon from Johns Hopkins Hospital in Baltimore used larval therapy to treat wounds for the treatment of osteomyelitis and gangrene wounds (41-43). Surgical researchers have known that larvae have some varieties of blow fly or green bottle after the 1700s (Phaenicia sericata and Lucilia sericata) and solely eliminate necrotic tissue, and repair the wound so the wound heals faster. With the discovery of penicillin by Flemming (1929) followed by Florey (1939) the use of larval therapy was abandoned. Due to antibiotic resistance and increasing chronic wound problems worldwide, larval therapy has been reintroduced and has been approved by the FDA for the use of aseptically produced larvae. Various advantages of larval therapy have been suggested and the main ones are debridement, ie removing necrotic tissue; enzymatic degradation of necrotic tissue; antimicrobial effect of E/S larvae; and stimulation of granulation tissue formation (Fig. 2) (6, 44).

Mechanical debridement

Blow fly larvae have a 10-14 day life cycle from hatching to adulthood. Medical larvae are produced in sterile conditions and because they are maintained in a moist state, these larvae do not pupate (45). These larvae also will not infiltrate the wound and damage healthy tissue because the larvae only degrade, liquefy, and ingest necrotic tissue. Various proteolytic enzymes have been identified from E/S larvae that can mechanically debridement and proteolytic digestion for 3-5 days can liquefy laminin and fibronectin from the extracellular matrix of necrotic tissue (46,47). Furthermore, the larvae quickly remove pathogens that enter their digestive tract, as well as promote and repair wound granulation tissue to accelerate wound healing (46-48). Larval therapy can suppress both formation and biofilms that have formed. The presence of larval movements (crawling and wandering) in the wound also affects the survival of bacteria, especially anaerobic bacteria that can form biofilms and are less resistant when passing through the larva's digestive tract (49). The larvae also secrete DNAse which degrades DNA in the exopolymeric matrix so that it can weaken the biofilm structure (6, 50).

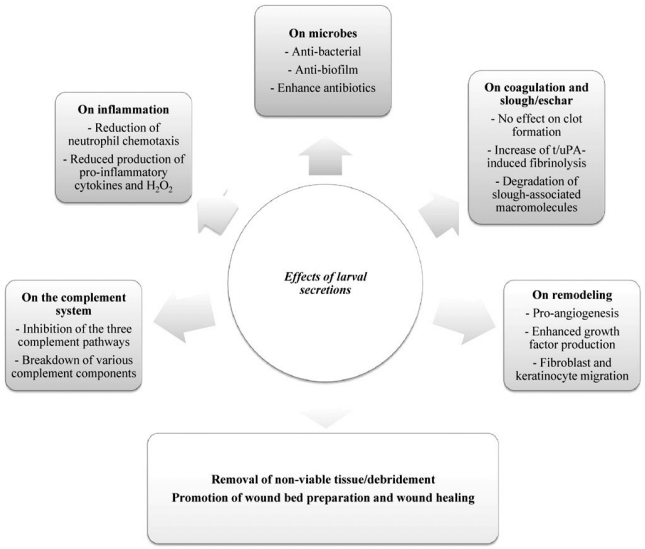

Fig. 2: An overview of the work of larval secrections in wounds (26).

DISCUSSION

Wound is a condition of damage to the continuity of tissue, anatomical structure and function of normal skin or mucosa due to pathological processes originating from the internal or external environment and affecting certain organs. Treatment and management of wounds in this case is one of the factors that determine the final outcome of the wound healing process (11). At the moment, various treatment methods have been studied for the treatment of various diseases (51, 52). One of the most important of these methods is 'Biotherapy". larval or Moggot therapy is a subset of biotherapy (53). Larval therapy is one of the modern methods of treatment in medicine and veterinary sciences. In this method, the use of sterile fly larvae is used to treat human and animal wounds. In the methodology of this treatment method, the larvae of Lucilia sericata (common green bottle fly) are used. The performance of modern larval therapy in the treatment of chronic wound and infectious wounds is outstanding. In some cases, larval therapy heals wounds faster and better than conventional methods (52,53).

Larval therapy is used in chronic wounds especially those that have failed conventional therapy. By developing bioactive molecules contained in larval secretion and excretion materials, it is hoped that larval therapy can be widely used to obtain more optimal results at a fairly economical cost (54).

CONCLUSION

Treatment of larvae is a method that is very easy to apply and is successful in traditional and complementary medicine. Although it has been discredited from time to time in the historical process, it has become the most widely used FDA-approved form of treatment today. The fact that this drug has almost no side effects, reduces the need for antibiotics and the results of successful studies make the treatment of larvae an important reason for preference in the treatment of chronic wounds. It is interesting to see success with MDT in the treatment of chronic wounds, especially in cases where conventional treatment has not been successful. The microdebridement ability and proteolytic enzymes of the larvae appear to be the key to this success.

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