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SKIN MICROBIOTA AND ITS INTERPLAY WITH WOUND AND BURN HEALING: IMPACT OF EPITHELIAL BIOREGENERATOR

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Abstract. *The skin microbiota is intimately coupled with cutaneous health and disease. Interactions between commensal microbiota and the multiple cell types involved in cutaneous wound healing regulate the immune response and promote barrier restoration. This dialog between host cells and the microbiome is dysregulated in chronic wounds and burns. To investigate whether changes in composition were present in the skin microbiome of individuals at risk of developing these lesions. Colonization of the wound and burn with commensal bacteria may promote wound and burn healing by inducing antimicrobial proteins such as Perforin-2, thus stimulating a protective immune response against pathogenic bacteria. Wound and burn infection with pathogenic bacteria results in Perforin-2 suppression in both hematopoietic and nonhematopoietic cells and inhibition of healing. A new study now shows that, in most cases, the causative agents of these infections are bacteria from the patient's own skin. For this reason, authors investigated the impact of Epithelial Bioregenerator to eliminate microorganisms from the chronic wounds and burns.*

Keywords: microbiome, host-pathogen interactions, chronic wounds, burns, infection, Epithelial Bioregenerator

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MATERIALS AND METHODS

Pathogenic microorganisms are suspected to play a substantial role in delayed wound and burn healing. These studies indicate that functional level differences between microbiota species, or even between specific microbiota strains, may play an important role in determining the clinical outcomes of chronic wounds and burns. Recent advances have enabled better characterization of bacteria in chronic wounds and burns.

RESULTS

We describe how, in contrast to pathogenic species capable of subverting skin immunity, commensals are essential for the regulation of the cutaneous immune system and provide protection from intracellular pathogens through modulation of the

antimicrobial molecule, Perforin-2. Thus, analyzing skin microbiota composition in both the normal acute and impaired wound and burn healing processes is indispensable for the identification of novel therapeutic strategies for patients with chronic wounds and burns. Perforin-2 has been shown to be critical for the clearance of a variety of Gram-positive and Gram-negative pathogens, highlighting its pivotal role in the host's innate antimicrobial response.

CONCLUSIONS

Chronic wounds and burns exhibit a hyperproliferative and non-migratory epidermis, unresolved inflammation, and fibrosis. Recent studies also showed that the skin microbiome is involved in wound and burn healing. Interactions between different species within the polymicrobial environment have been shown to be dynamic and to modify bacterial behavior, resulting in increased virulence and delayed wound and burn healing. It is necessary to further investigate such microbiome-host interactions to identify new potential treatments.

Background

The skin, the largest organ of the human body, consists of several layers (epidermis, dermis and hypodermis) which, together, have an important role in preventing the penetration of microorganisms and potential toxins and in preventing infection, while controlling mode. time loss of water and nutrients. Above the epidermis there is an aqueous and lipid layer that contributes to the ecology of the surface. At the forefront of this process of maintaining homeostasis is the epidermal barrier, with the main type of cells - keratinocytes, which are the first active participants in the host's immune response. The skin, the largest human organ, is colonized by billions of microorganisms, with a great variety (bacteria, fungi, viruses, protozoa), which have adapted to the chemical and physical characteristics of the skin and formed the skin microbiome or microbiota. Staphylococcus epidermidis and other coagulase-negative staphylococci have been considered the main colonizers of the skin. With the advent of molecular technologies for the detection and identification of microbial genes, it has become apparent that the germ growth technique detects only a small fraction of the total skin microorganisms. After the gut, the most numerous microorganisms are located in the skin.

The physiology of wound healing under normal conditions is achieved via several defined steps: inflammation, proliferation, epithelialization, maturation, and remodeling phases (1).

The skin microbiota of healthy adults remains stable over time, despite environmental perturbations-at least on earth. It is well known that skin diseases and disorders, are associated with an altered microbial state. The skin microbiota has been studied by using both cultivable microbes and metagenomic profiles. The skin microbiome is protective against pathogens, nevertheless in certain conditions the

microorganisms that are ordinarily beneficial to their host can become pathogenic (dysbiosis). Skin microorganisms are important in educating the innate and adaptive arms of the cutaneous immune system, and the commensal skin microbiome during healing is essential for the regulation of the cutaneous immune system. Under certain conditions, skin microorganisms that are normally beneficial to their host can become pathogenic. When an imbalance occurs between commensal and pathogenic agents, a skin disease or even a systemic disease can result. Many common skin diseases are associated with changes in the balance of the microbiota, called dysbiosis. This dysbiosis is often caused by common commensal species, as described below for dermatitis, acne, chronic wounds, etc.

Currently, few details exist regarding the description and function of non-bacterial commensal germs. At the skin level, part of the microbiota is common to all humans, while another part is unique to each individual, this diversity being influenced by a number of factors such as skin anatomy, host-related factors, environmental factors, etc. the microbiome plays a critical role in protecting against invading pathogens and educating our immune system, but under certain circumstances, when the balance of this microbial community is disrupted, some skin commensal germs can become pathogens, causing a skin disease or even a systemic. Skin microorganisms can provide protection from pathogens through modulation of antimicrobials (2).

There are many unknown aspects of the skin microbiome and its role in human health and pathology that require further in-depth study. there is little detail regarding the description and function of non-bacterial commensal germs. At the skin level, part of the microbiota is common to all humans, while another part is unique to each individual, this diversity being influenced by a number of factors such as skin anatomy, host-related factors, environmental factors, etc. the microbiome plays a critical role in protecting against invading pathogens and educating our immune system, but under certain circumstances, when the balance of this microbial community is disrupted, some skin commensal germs can become pathogens, causing a skin disease or even a systemic.

The role of the microbiome in chronic wound is not fully understood and the importance of topical antimicrobial agents in their treatment is continuously debated. Pathogens or pathobionts are suspected to delay the healing process (3).

Microorganisms can act competitively to exclude one another or synergistically for mutual benefits depending by the interaction between the skin surface and other microorganisms that live on it. The immune system is interconnected with the skin microbiota, especially by targeting pathogen-associated molecular patterns (PAMPs), through pattern recognition receptors (PRRs). For example, Propionibacterium acnes and the lipopolysaccharides induce the expression of antimicrobial peptides and proinflammatory cytokines/chemokines in human sebocytes contributing to the host defense and skin inflammation (4).

Microorganisms could be beneficial if able to regulate the immune response toward a normal healing process. For example, the lack of interleukin-10 (IL-10) (a key anti-inflammatory cytokine in immunologic tolerance) results in a strong inflammatory response. IL-10 is a key mediator of the pro-to anti-inflammatory transition that counters collagen deposition (5).

In healthy individuals, IL-10 expression was positively correlated with the abundance of the gammaproteobacterial genus Acinetobacter on the skin. The microbial populations settled on skin, space modules, in space suits, are also playing a pivotal role, as wound healing is also affected by the microbial community (6).

The natural antimicrobial properties of the skin microbiota, the crosstalk of the skin microbiota with the immune system during wound healing, the contribution of the microbiota in precision medicine, and the role of gut-skin and gut-brain axes (7).

Commensal bacteria can modulate the cutaneous immune response to prevent wound infections. Cutaneous wound healing is a complex and very organized process, tightly controlled by several cell types through the secretion of growth factors, cytokines, and chemokines (8,9).

Disruption of this process prevents the skin barrier from being properly restored; thus, the wound does not heal and becomes chronic. Higher rates of keratinocyte proliferation, an absence of migration, and fibrosis have all been observed in chronic wounds, regardless of their type. In contrast to the normal process of wound healing, angiogenesis, stem cell recruitment and activation, and extracellular matrix remodeling have all been shown to be impaired in chronic wounds, whereas inflammation has been found to be persistent and unresolved (10).

Colonization of wounds with commensal microbiota may promote wound healing through activation of the innate immune response (11).

In the proliferation phase, keratinocytes multiply and migrate, fibroblasts migrate and deposit extracellular matrix, and angiogenesis occurs. Wounds provide an opportunity for microorganisms from the skin surface that constitute the skin microbiota, coupled with those from the environment, to gain entry to the underlying tissues and find optimal conditions for colonization and growth. Interaction of commensal microorganisms with the skin cells during the normal cutaneous wound healing process is thought to be beneficial in modulating the innate immune response (12,13). The microbial typing for each individual is ideal for precision medicine approaches, leading treatment of wounds. In this context it would be interesting to stimulate ad hoc skin probiotic communities in an ecology-based therapy scenario, by limiting dysbiosis that leads to cutaneous disorders (14)

The skin microbiota is intimately coupled with cutaneous health and disease. Interactions between commensal microbiota and the multiple cell types involved in cutaneous wound healing regulate the immune response and promote barrier restoration. This dialog between host cells and the microbiome is dysregulated in chronic wounds and burns (15).

With the appearance and development of molecular methods, it was found that the skin hosts a much greater variety of microorganisms, generally considered as colonizers of the skin.

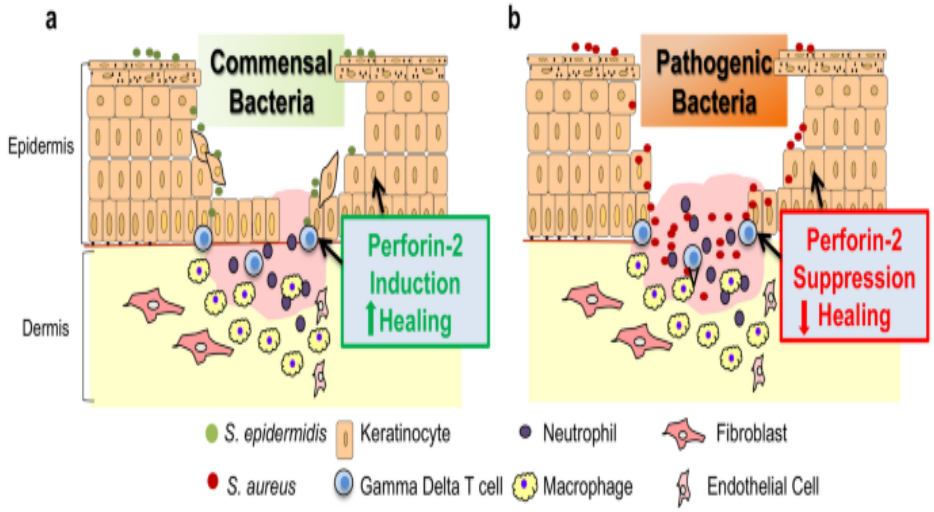
The detection and characterization of skin microbes were initially carried out by the method of cultivation on culture media of samples collected from the skin with the help of swabs. With the advent of molecular technologies for the detection and identification of microbial genes, it became clear that the culture technique detects only a small part of the total microorganism nisms that interact on the surface. DNA sequencing techniques have identified and described the diversity of resident microbes inside our bodies, and thus, while the term "biome" is used to describe the ecology of the human body, the microbial communities of the human body have been called the" human microbiome". To understand the skin microbiota, it is essential to recognize that, unlike all other frequently studied areas of the human microbiome, such as the gut and oral mucosa, the skin has the greatest diversity of microorganisms.

To investigate whether changes in composition were present in the skin microbiome of individuals at risk of developing these lesions, Redel et al. conducted a case-control observational study using high-throughput 16S rRNA sequencing technology to analyze the skin microbiota from the arms and feet of diabetic men without any previous history of diabetic foot ulcers and from those of matched nondiabetic men as controls (16). Although microbiota composition and total bacterial counts were similar in the arm samples of both groups, higher bacterial diversity was observed in the plantar foot samples of diabetic men compared to nondiabetic men (17).

The term" microbiota" is also used to describe these microorganisms and specifically means:" the ecological community of commensal, symbiotic, and pathogenic microorganisms that can literally exist in our body space".

Cutaneous immunity is differentially regulated by commensal and pathogenic microorganisms through modulation of perforin-2 (5)

There are many unknown aspects of the skin microbiome and its role in human health and pathology that require further in-depth study. Unlike the knowledge about the role of the intestinal microbiota, the knowledge about the microbiome of the human skin is much less. Unfortunately, not many details are currently known regarding the description or functions of the nonbacterial commensal microbes of the skin. Worldwide, more studies are needed to clarify the role of these microbial communities, both in terms of human health and pathology.



Objective To investigate whether changes in composition were present in the skin microbiome of individuals at risk of developing these lesions. Colonization of the wound and burn with commensal bacteria may promote wound and burn healing by inducing antimicrobial proteins such as Perforin-2, thus stimulating a protective immune response against pathogenic bacteria. Wound and burn infection with pathogenic bacteria results in Perforin-2 suppression in both hematopoietic and nonhematopoietic cells and inhibition of healing. A new study now shows that, in most cases, the causative agents of these infections are bacteria from the patient's own skin. For this reason, authors investigated the impact of Epithelial Bioregenerator to eliminate microorganisms from the chronic wounds and burns.

Materials and methods Infection remains a leading cause of mortality for burn patients, despite advances in burn wound management that have improved outcomes in recent years. Early excision and grafting of burn eschar, advanced dressings, and topical antiseptics and antimicrobials play an important role in preventing infection, but systemic antimicrobial therapy remains a relatively common necessity when infection overwhelms local defenses. Burn patients are at high risk of sepsis due to a number of factors: damage to the epithelium leading to colonization of the wound with pathogens; local loss of local immune cells and innate immune proteins through fluid and protein loss; insertion of intrusive devices such as catheters and breathing tubes; and the hypermetabolic state leading to further loss of proteins involved in innate immune responses and neutrophil dysfunction.

Studies from the 1950s to the 1990s have demonstrated a different era of burn care in which burn-related morbidity and mortality rates, as well as secondary infections such as sepsis, were much higher than today's rates. This was the result of bacterial infection and colonization of the burn wound surface that was suppressed but not completely eliminated. However, this era has shaped our understanding of

burn wound epidemiology over time. Pathogenic microorganisms are suspected to play a substantial role in delayed wound and burn healing. These studies indicate that functional level differences between microbiota species, or even between specific microbiota strains, may play an important role in determining the clinical outcomes of chronic wounds and burns. Recent advances have enabled better characterization of bacteria in chronic wounds and burns.

Results Infection arising from burn wounds may occur early or after closure and may be associated with debridement procedures. Clinical examination of wounds is heavily relied upon to determine whether wounds are a source of infection, as burn wounds frequently cultivate pathogens that often colonize flora from the natural microbiome of the skin, however, colonizing pathogens can cause biofilms and infections in the local area of the burn injury. Increased volume of exudate, development of purulent exudate, epithelial loss or graft loss, local cellulitis, sinus tract or abscess formation are signs of local burn wound infections. Local wound infections delay wound healing due to the presence of over-colonizing pathogens that develop an extracellular matrix on the wound surface. The pathogenic extracellular matrix inhibits re-epithelialization and maintains the wound in an increased inflammatory state due to the host's response to the presence of the pathogen (3). The consequence of this delay affects the course of injury, directly affecting the outcomes of wound healing, scarring, and any hypermetabolic response associated with injury.

If left to persist, local infections lead to systemic infections with severe consequences for wound healing and mortality rates, demonstrated by a mortality rate of twice that in burn patients compared to uninfected patients. Burn wound infections and sepsis are important complications, as 42% to 65% of burn deaths have been shown to be associated with infection, depending on the size and severity of the body burn injury (11). In severe cases involving invasive devices required to support the patient, including intubation and catheters, and prolonged hospitalization, it predisposes patients to subsequent infections.

The most common infectious syndromes in burn patients are complications arising from local infections and skin grafts at the site of the burn injury, but infections may also occur and may be associated with pneumonia and urinary tract infections (5). Pneumonia including ventilator-associated pneumonia in the early phase of burns is particularly common in those with inhalation burns (13), with reports indicating the combination of fire injury and more than four days of mechanical ventilation increases the frequency of pneumonia (17). Blood and urinary tract infections from catheter use can also occur at any time. Sepsis remains the leading cause of death after the first 24 hours of admission to a burn hospital and persists as a risk even after the first two weeks of admission (12). Surveillance wound cultures should be performed regularly. In addition, regular screening of nasal, groin, and axillary swabs for methicillin-resistant Staphylococcus aureus (MRSA) and rectal swabs for vancomycin-resistant Enterococcus (VRE) and multidrug-resistant Gram-

negative bacteria should be performed to guide empiric antimicrobial therapy if sepsis develops.

We describe how, in contrast to pathogenic species capable of subverting skin immunity, commensals are essential for the regulation of the cutaneous immune system and provide protection from intracellular pathogens through modulation of the antimicrobial molecule, Perforin-2. Thus, analyzing skin microbiota composition in both the normal acute and impaired wound and burn healing processes is indispensable for the identification of novel therapeutic strategies for patients with chronic wounds and burns. Perforin-2 has been shown to be critical for the clearance of a variety of Gram-positive and Gram-negative pathogens, highlighting its pivotal role in the host's innate antimicrobial response (18).

The pathology of wound healing is now well characterized, the cellular and molecular mechanisms of impaired wound healing are still being studied. Interaction of commensal microorganisms with the skin cells during the normal cutaneous wound healing process is thought to be beneficial in modulating the innate immune response (19). Conversely, pathogenic microorganisms are suspected to play a substantial role in delayed wound healing. A complex microbiome is a hallmark of chronic nonhealing wounds (20). Pathogenic bacteria are able to escape skin immunity and even reside host cells. Thus, analyzing skin microbiota composition in both the normal acute and impaired wound healing processes is indispensable for the identification of novel therapeutic strategies for patients with chronic wounds. Probiotics actively can crosstalk between the immune system and the skin microbiota.

Epithelial bioregenerator Deniplant

Presentation In the form of a concentrated solution.

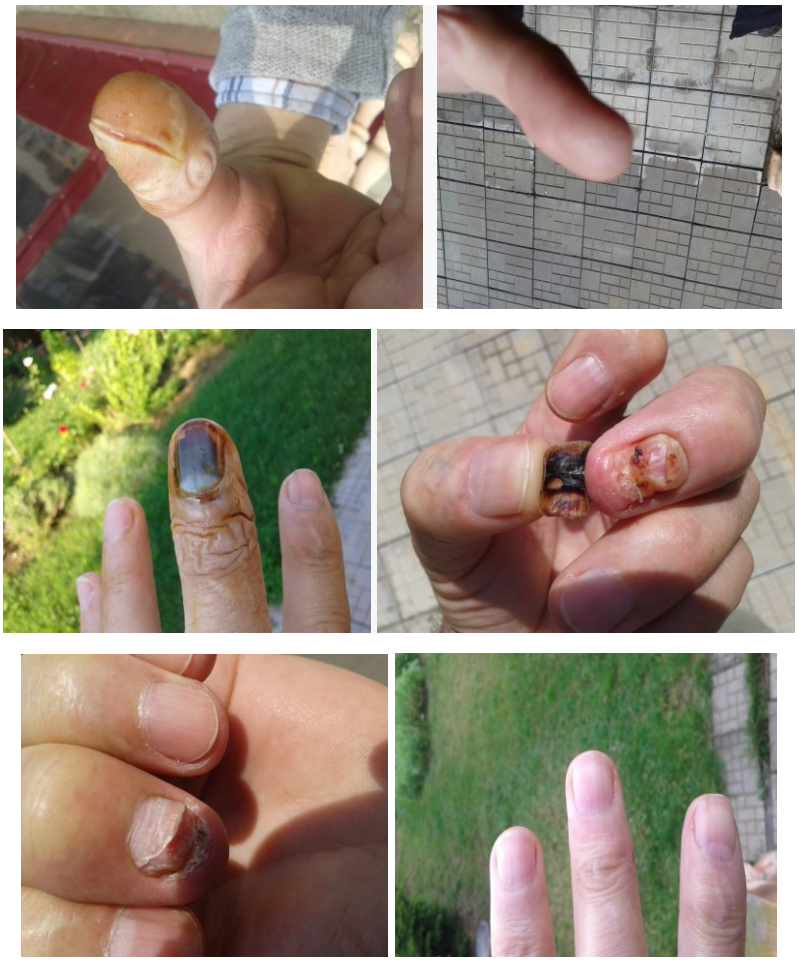
Action, It has intense anti-inflammatory, antiseptic, antipruritic, anti-algesic, hemostatic action; facilitates the penetration of active substances into the skin.

Indications Cut injuries, open wound, stops bleeding, isolates the injury, relieves stinging and pain. In traumatic skin injuries (open injuries, wounds) it stops bleeding very quickly, prevents infection, prevents skin necrosis, restores the destroyed tissue very quickly, the traces of the injury disappear in a short time, in superficial injuries no dressing is necessary, the injury is kept clean, making cleaning easier. In burns of various degrees, the sting and pain disappear, restores the destroyed tissue very quickly, if pustules have formed, they retreat, prevents infection and skin necrosis, no occlusive dressing is necessary, the injury is kept clean, making cleaning easier. In cosmetic treatments: prevents and removes acne, stubborn pimples, heals damaged tissue without leaving visible traces, when subdermal problems occur, stops the inflammatory process, restores inflamed eye conjunctival tissue. In plastic and reparative surgery, it favors the rapid recovery of the grafted tissue, excludes the possibility of infection. Against wasp, bee or spider stings.

Administration It is applied to the lesion repeatedly, after penetrating the skin, until the stinging and pain disappear, then infrequent applications (3-4 times) a day until complete healing. After a while when the sting or pain appears, it must be put on the wound again.

A paper soaked in the solution can also be placed for insulation or the solution can be kept on the lesion for several minutes (30 minutes). After cleaning the wound with disinfectant, apply the solution again. No occlusive dressing is required. If it is washed with water, the solution must be applied again for a few minutes. The burn is covered with the solution until the sting disappears and the burn is isolated. The solution is applied to the wounds until the pain disappears, then it is repeated so as not to blacken the skin.

How to store Store in a cool, dark place.



CONCLUSIONS

Most studies have focused on common burn pathogens, including P. aeruginosa and K. pneumonia, however, burn care combats a wide range of pathogens, including fungi and multi-resistant strains, that require attention additional. Chronic wounds and burns exhibit a hyperproliferative and non-migratory epidermis, unresolved inflammation, and fibrosis. Recent studies also showed that the skin microbiome is involved in wound and burn healing. Interactions between different species within the polymicrobial environment have been shown to be dynamic and to modify bacterial behavior, resulting in increased virulence and delayed wound and burn healing. It is necessary to further investigate such microbiome-host interactions to identify new potential treatments.

Statement of Competing Interests

The authors declare that they have no known competing financial interests or personal relationships that could appear to influence the work reported in this paper.

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