ACTIVATED COLLAGEN ACCELERATES WOUND REPAIR AND MODULATES CYTOKINE PRODUCTION IN WHOLE BLOOD AND PBMC CULTURES.

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Abstract

Several reports have shown that enzymatically activated collagen fragments enhance wound repair through poorly understood mechanisms. CellerateRX is an activated (fragmented) product derived from Type I Collagen that is applied topically as a gel or powder on open wounds that are often infected or colonized with bacteria. In these case series, topical CellerateRX in conjunction with standard care healed infected surgical wounds and diabetic ulcers more rapidly than standard therapy alone. We conducted an in vitro analysis of 16 patients with lower extremity diabetic ulcers. Eight patients received CellerateRX in addition to standard Treatment, and 8 patients received standard treatment alone. A quantitative analysis of wound healing at 14 days showed that CellerateRX-treated wounds were 100% healed, compared to 59%±15% healing in control wounds. To examine possible mechanisms by which CellerateRX enhances wound healing, cytokine production was evaluated in whole blood from diabetic patients treated with CellerateRX. CellerateRX induced IL-1β, TNF-α, and IL-10 synthesis was not significantly affected by any CellerateRX concentration tested.

Background

The acute immune response is activated in wounds and includes cytokine biological effects, which are rapid and highly regulated. Cytokine activities important for physiological wound healing include effects on coagulation, inflammation, epithelialization, angiogenesis, matrix and tissue remodeling, and pathogen control. CellerateRX, composed primarily of proteolytically cleaved collagen, is thought to accelerate wound healing by incompletely understood mechanisms. We investigated the effect of CellerateRX in a clinical investigation of wound healing, and we assessed CellerateRX effects on pre- and anti-inflammatory cytokines thought to be relevant in wound repair and pathogen control.

Methods

Wound healing in diabetic patients: Sixteen patients with below-knee diabetic ulcers were recruited. Eight patients (Controls) were treated with standard therapy, consisting of an antibiotic occlusive dressing secured with an elastic strap. Eight patients (CellerateRX) were treated with a 10-15 g CellerateRX powder (Wound Care Innovations, LLC, Florida) in addition to the standard therapy. Wounds were treated and photographed weekly, and wound areas were quantified using image analysis.

Whole blood assays: Heparinated human blood was obtained from 10 healthy volunteers and diluted 1:5 with RPMI tissue culture medium in the absence (CellerateRX=0) or presence of CellerateRX. After 1 hr of incubation at 37˚C, cultures remained unstimulated, or were stimulated with 1.2 µg/ml (200 pg/ml) heat-killed Staphylococcus aureus (S.A.) or lipopolysaccharide (LPS). In whole blood, CellerateRX significantly increased spontaneous production of IL-1β, TNF-α, IL-6, IL-10, tumor necrosis factor alpha (TNF-α), and IL-10. In whole blood stimulated with S.A. cytokine synthesis was augmented by CellerateRX, except for TNF-α, which was suppressed. In PBMC cultures, CellerateRX significantly induced spontaneous production of IL-1β, IL-6, IL-10, TNF-α, IL-10, and IL-10. CellerateRX stimulation with LPS, CellerateRX significantly augmented IL-1β, IL-6, and IL-10. TNF-α was suppressed. CellerateRX inhibition of stimulated TNF-α in whole blood and PBMC suggests selective suppression of detrimental TNF-α inflammatory and cell death effects in vivo. These results suggest that CellerateRX enhances wound healing, and a possible mechanism involves specific modulation of the cytokine response in bacteria-containing wounds.

Results

A. Clinical Wound Healing

1. Treatment of diabetic ulcers with CellerateRX and standard therapy significantly accelerated wound healing compared to standard therapy alone.

B. Whole Blood Culture Cytokines

1. CellerateRX significantly increased spontaneous (unstimulated) levels of IL-8, IL-6, and IL-10. A biphasic CellerateRX effect on TNF-α and IL-1β was observed, with stimulation at lower CellerateRX concentrations and no effect at higher concentrations. CellerateRX treatment of patients had a mean±SEM percent wound healing of 50%±15, and a median value of 100. *P<0.002 by Mann-Whitney test. Horizontal bars indicate median values. B. Representative photogaphs of lower extremity ulcers on patients treated with standard therapy (left panels) or treated with standard therapy plus CellerateRX (right panels).

Discussion

Discussion

Diabetes-related lower extremity infections are a major clinical problem. In the U.S., 6–10% of all diabetic patients experience lower extremity ulcer infections, which result in over 50,000 amputations each year. Lower extremity diabetic ulcer care continues to be complicated by the emergence of antibiotic-resistant bacteria. Although it is reasonable to assume that CellerateRX is an activated (fragmented) product derived from Type I Collagen that is applied topically as a gel or powder on open wounds that are often infected or colonized with bacteria. In these case series, topical CellerateRX in conjunction with standard care healed infected surgical wounds and diabetic ulcers more rapidly than standard therapy alone. We conducted an in vitro analysis of 16 patients with lower extremity diabetic ulcers. Eight patients received CellerateRX in addition to standard Treatment, and 8 patients received standard treatment alone. A quantitative analysis of wound healing at 14 days showed that CellerateRX-treated wounds were 100% healed, compared to 59%±15% healing in control wounds. To examine possible mechanisms by which CellerateRX enhances wound healing, cytokine production was evaluated in whole blood from diabetic patients treated with CellerateRX. CellerateRX induced IL-1β, TNF-α, and IL-10 synthesis was not significantly affected by any CellerateRX concentration tested. CellerateRX did not affect LPS-stimulated IL-1β production and significantly decreased LPS-stimulated levels of TNF-α.