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Review

TWO₂ Therapy Demonstrates Clinically Meaningful Long-Term Outcomes Compared to Other Advanced Wound Care Modalities: Real-World Evidence Supported by Mechanistic and RCT Clinical Data

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Abstract

Background/Objectives: Chronic diabetic foot ulcers (DFUs) and venous leg ulcers (VLUs) remain a major source of morbidity, healthcare utilization, and limb loss, despite adherence to established standards of care protocols and the widespread availability of advanced wound technologies. Many advanced modalities only target isolated aspects of wound healing and fail to address the complex, interdependent pathophysiology of chronic wounds, particularly tissue hypoxia, edema, impaired microcirculation, and persistent inflammation. Cyclical Pressurized Topical Wound Oxygen (TWO₂) therapy is a home-based, multimodal intervention that combines humidified topical oxygen delivery with cyclical non-contact compression to address these core drivers simultaneously. **Methods:** This review synthesizes mechanistic rationale and evidence from randomized controlled trials, long-term venous ulcer studies, and real-world comparative effectiveness analyses. Emphasis is placed on the large cohort study by Yellin et al., which directly compared TWO₂ with other advanced modalities including negative pressure wound therapy (NPWT), skin substitutes, and growth factor therapies. **Results:** Across these studies, TWO₂ therapy is consistently associated with improved healing durability, reduced recurrence, and substantial reductions in hospitalization and amputation rates compared with both standard care and advanced wound therapies. **Conclusions:** The convergence of randomized and real-world evidence supports TWO₂ therapy as a clinically meaningful and mechanism-driven adjunctive treatment option for patients with chronic, high-risk lower-extremity wounds.

Keywords: chronic wounds; topical oxygen therapy; intermittent compression; cyclical pressurized topical wound oxygen (TWO₂); multi-modality intermittent topical oxygen therapy (ITOT); inflammation resolution; wound healing; advanced wound care



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1. Introduction

Chronic lower-extremity wounds represent a growing global health challenge, driven by the increasing prevalence of diabetes, vascular disease, and advanced age [1]. Diabetic foot ulcers (DFUs) and venous leg ulcers (VLUs) are among the most challenging to manage, frequently resulting in serious complications such as infection, hospitalization, and amputation. These adverse outcomes contribute significantly to morbidity, mortality, and healthcare costs [2–5]. Despite the availability of numerous advanced wound care technologies, real-world outcomes remain suboptimal. Many wounds fail to progress beyond the inflammatory phase of healing, even when treated with advanced therapies, such as negative pressure wound therapy, bioengineered skin substitutes, or topical growth factors. A common limitation of these approaches is their inability to adequately correct the chronic wound microenvironment, which is characterized by hypoxia, edema, impaired microcirculation, and excessive protease activity [6–8]. Cyclical Pressurized Topical Wound Oxygen (TWO₂) therapy has emerged as a promising adjunctive treatment modality capable of directly addressing oxygen deficiency while concurrently reducing edema, improving perfusion, and modulating inflammation.

This article synthesizes findings from randomized and real-world studies, contextualizes outcomes against other advanced treatments, and integrates mechanistic insights that provide the foundation for the clinically meaningful long-term healing outcomes observed with TWO₂ therapy.

2. Materials and Methods

This is a narrative review of the mechanistic, randomized, and real-world evidence base for TWO₂ therapy in chronic lower-extremity wounds. Literature searches were conducted in PubMed/MEDLINE (National Library of Medicine, Bethesda, MD, USA; <https://pubmed.ncbi.nlm.nih.gov>, accessed on 1 April 2026), the Cochrane Library (Cochrane, London, UK; <https://www.cochranelibrary.com>, accessed on 1 April 2026), and [ClinicalTrials.gov](https://clinicaltrials.gov) (U.S. National Institutes of Health, Bethesda, MD, USA; <https://clinicaltrials.gov>, accessed on 1 April 2026) for studies published between January 2000 and March 2026, using the search terms: diabetic foot ulcer, venous leg ulcer, topical oxygen therapy, TWO₂, ITOT, intermittent topical oxygen, wound healing, negative pressure wound therapy, hyperbaric oxygen therapy, skin substitutes, growth factors, and chronic wound. Inclusion criteria were peer-reviewed human studies in the English language with relevance to mechanistic rationale, comparative effectiveness, or long-term clinical outcomes. Studies were selected by the authors based on clinical relevance, methodological quality, and contribution to the evidence base. References were organized and managed using Mendeley Reference Manager (version 2.144.0; Elsevier, Amsterdam, The Netherlands; <https://www.mendeley.com>, accessed on 1 June 2026).

2.1. Pathophysiologic Rationale for Oxygen-Based Multimodal Therapy

Chronic wounds differ fundamentally from acute wounds in both biology and behavior. Rather than progressing through the orderly phases of healing, chronic wounds remain trapped in a state of persistent inflammation [9–11]. Tissue hypoxia is a defining feature of this state, resulting from macrovascular disease, microvascular dysfunction, edema-related capillary compression, impaired oxygen diffusion, and inflammatory metabolic demand [12].

Hypoxia disrupts multiple healing pathways. Fibroblast proliferation and collagen deposition are oxygen-dependent processes, as is angiogenesis. Immune function is similarly compromised, reducing bacterial killing, and increasing the risk of infection [12].

Edema increases interstitial pressure, compresses capillaries, and lengthens oxygen diffusion distance, further amplifying tissue hypoxia. Inflammatory mediators and proteases accumulate due to inadequate lymphatic clearance, degrading growth factors and extracellular matrix components [13,14]. Repetitive ischemia–reperfusion injury generates oxidative stress, further impairing tissue repair [6].

Effective chronic wound therapy must therefore address oxygen delivery, fluid dynamics, edema, and inflammation in an integrated manner. Therapies that target only wound size, exudate, or surface coverage may fail to produce durable healing if these underlying drivers are not corrected.

2.2. The Synergy of Topical Oxygen, Cyclical Compression and Humidification

Cyclical Pressurized Topical Wound Oxygen therapy was developed to address the multifactorial pathophysiology of chronic wounds through a single, integrated system. The therapy combines three synergistic components: topical oxygen delivery, cyclical non-contact compression, and humidification.

Topical oxygen delivery increases tissue oxygen tension at the wound surface, supporting oxidative bacterial killing, collagen synthesis, and angiogenesis [12,15–20]. Unlike systemic oxygen approaches, topical delivery targets the wound directly, minimizing systemic exposure while maximizing local effect [16].

Therapeutic level cyclical compression augments these effects by increasing the oxygen partial pressure delivered to the wound bed, facilitating lymphatic clearance of inflammatory mediators, reducing edema, restoring microvascular pressure gradients, and improving oxygen diffusion into hypoxic tissue [6]. At the cellular level, cyclical mechanical deformation activates endothelial and stromal mechanotransduction pathways, including eNOS and integrin-mediated signaling, that promote inflammation resolution, angiogenesis, fibroblast proliferation, extracellular matrix deposition, and collagen cross-linking [6] (Figure 1).

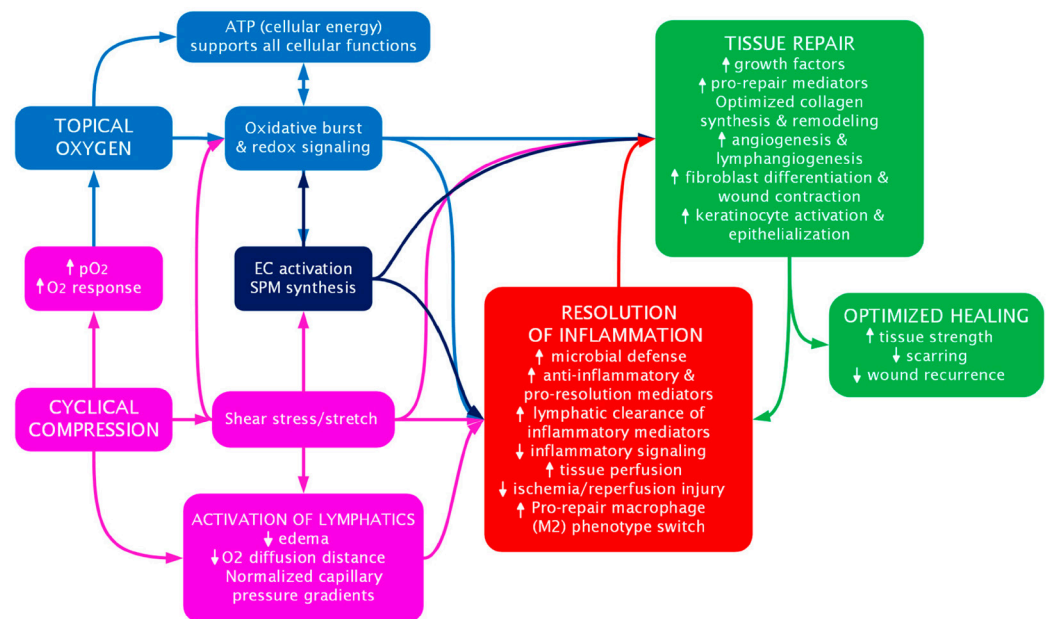


Figure 1. TWO₂ Mechanism of Action: This figure presents a conceptual framework of the proposed integrated cellular and molecular mechanisms by which topical oxygen and cyclical compression synergistically promote wound healing. Pathways shown are derived from published mechanistic and clinical literature; the figure is not intended to depict quantitative relationships, and the modulating influence of variables such as wound age, severity, and comorbidity burden is recognized. Topical oxygen increases tissue oxygen tension to fuel ATP production, enhance microbial defense via oxidative

burst, activate redox signaling, and optimize collagen synthesis and crosslinking. Cyclical compression increases the partial pressure (pO_2) of topical oxygen and activates lymphatic function, improving clearance of inflammatory mediators, reducing edema, decreasing diffusion distance for oxygen, normalizing capillary pressure gradients, and restoring perfusion. Compression-induced shear stress and shear stretch activate endothelial cells (ECs) and stimulate the biosynthesis of specialized pro-resolving mediators (SPMs). This initiates a cascade of anti-inflammatory and pro-resolution signaling, including polarization of macrophages toward the reparative M2 phenotype and acceleration of inflammation resolution. In parallel, M2 macrophages, SPMs, and ECs upregulate growth factors and reparative cytokines that direct wound repair and remodeling. Activated ECs stimulate angiogenesis and lymphatic angiogenesis, while fibroblasts drive collagen synthesis, ECM production, and myofibroblast differentiation, enabling wound contraction. Keratinocyte activation promotes epithelialization, and during remodeling, fibroblast activity enhances collagen fiber organization. Collectively, these pathways promote efficient resolution of inflammation, improved perfusion, increased tissue strength, reduced scarring, and lower wound recurrence. pO_2 , partial pressure of oxygen; EC, endothelial cell; SPM, specialized pro-resolving lipid mediators; NADPH, nicotinamide adenine dinucleotide phosphate; ATP, adenosine triphosphate; M2, pro-repair macrophage phenotype (Illustration by Blakely MM, 2026, reprinted/adapted from ref. [6]).

Humidification improves oxygen diffusion, maintains a moist wound environment, and supports organized tissue formation and durable healing [21]. Together, these mechanisms promote a biologically favorable wound environment that supports not only closure but also durable tissue repair [6].

3. Results

3.1. Evidence from Randomized Controlled Trials

High-quality randomized evidence supporting TWO_2 therapy is provided by the multicenter, double-blind, sham-controlled TWO_2 Study conducted in patients with chronic DFUs that had failed to respond to standard care and published in *Diabetes Care* [22]. Participants were randomized to receive either active TWO_2 therapy or sham treatment in addition to optimal wound care.

At the first prespecified interim analysis, healing at 12 weeks was significantly higher in the active TWO_2 group compared with sham (41.7% versus 13.5%), with an adjusted odds ratio of 6.00 (97.8% CI 1.44, 24.93), $p = 0.004$, after controlling for ulcer severity. Time-to-healing analysis demonstrated a more than fourfold greater likelihood of healing over 12 weeks in the TWO_2 group (HR = 4.66 [97.8% CI 1.36, 15.98], $p = 0.004$).

Importantly, durability of healing was confirmed at one year, with a significant 56% of ulcers in the active group remaining closed compared with just 27% in the sham group ($p = 0.013$). This sustained healing at one year suggests improved tissue remodeling, which was further demonstrated by a sixfold reduced ulcer recurrence.

3.2. Evidence in Venous Leg Ulcers

In a prospective controlled study of 132 patients with chronic VLUs present for more than two years, TWO_2 therapy was compared to conventional compression dressings [23,24]. Key and statistically significant findings:

- Healing rate: 76% vs. 46% at 12 weeks ($p < 0.0001$)
- Median time to closure: 57 vs. 107 days ($p < 0.0001$)
- Recurrence at 36 months: 6% vs. 47% ($p < 0.0001$)

Pain reduction and improved infection resolution were also observed in the TWO_2 group. These findings extend the relevance of TWO_2 beyond diabetic wounds to those with venous pathology, where edema and hypoxia are central features. This is partic-

ularly important as most patients treated in the real world are comorbid with multiple etiological traits.

3.3. Real-World Evidence in Chronic Lower Extremity Wounds

While randomized trials establish efficacy, real-world evidence is critical to understanding clinical value across heterogeneous, high-risk populations. A recent large cohort retrospective study by Lohr et al., evaluated the effectiveness of TWO₂ in chronic lower extremity wounds of varying etiologies [25]. In this group of 3126 patients, 64.8% ($n = 2027$) achieved complete healing in 4.2 (SD ± 2.5) months, despite a mean pre-treatment wound age of 7 (± 15.9) months. The need for retreatment due to wound recurrence was only 2.7% ($n = 54$), with a mean follow up time of 13.9 (± 4.9) months and the rates of hospitalization and amputation were 3.7% ($n = 115$) and 6.1% ($n = 191$), respectively, substantially lower than historical standards [1,26–28].

Subgroup analyses demonstrated healing rates of 63.3% in a mean time of 4.2 (± 2.5) months in DFUs, 72.1% in a mean time of 4 (± 2.6) months in VLUs, 59.3% in a mean time of 3.9 (± 2.5) months in arterial ulcers, and 65.6% in a mean time of 4 (± 2.3) months in atypical wounds. Compared with large wound registry data, the more medically complex patients in the TWO₂ study had higher rates of healing in both DFU and VLU (63% at 18 weeks vs. 45% at 20 weeks for DFU, and 72% at 17 weeks vs. 57% at 16 weeks for VLU) [29]. The findings of this large study further support the efficacy of TWO₂ in chronic wounds of varied etiologies.

3.4. Real-World Comparative Effectiveness Evidence

Comparative effectiveness evidence is crucial to evidence-based clinical decision-making, empowering clinicians and patients to select treatments most likely to produce meaningful outcomes, and enabling health systems to direct resources toward interventions that demonstrate durable value. A retrospective cohort analysis of 202 patients by Yellin et al., evaluated the effectiveness of home-based TWO₂ therapy compared to other advanced therapies in patients with DFUs treated at two U.S. Veterans Affairs hospitals [30].

3.4.1. TWO₂ vs. No TWO₂ (Additive Model)

In this comparison, outcomes in patients who had received TWO₂ therapy at any point during their care pathway (TWO₂) versus those who had not received TWO₂ therapy (NO TWO₂) were evaluated. Patients in both groups may have received other additional advanced treatment interventions, therefore TWO₂ therapy was considered an additive adjunctive treatment.

In unmatched cohorts, patients treated with TWO₂ experienced 88% fewer hospitalizations (6.6% vs. 54.1%, $p < 0.0001$) and 71% fewer amputations (12.1% vs. 41.4%, $p < 0.0001$) over one year when compared with patients who did not receive TWO₂. After propensity score matching for age, wound severity, comorbidities, prior amputation, and use of other advanced therapies, TWO₂ therapy was associated with an 82% reduction in hospitalizations (7.1% vs. 40.0%, $p < 0.0001$) and a 73% reduction in amputations (8.6% vs. 31.4%, $p = 0.0007$), compared with no TWO₂. Logistic regression demonstrated an almost ninefold greater risk of hospitalization and fivefold greater risk of amputation among patients who did not receive TWO₂. These reductions persisted across Wagner grades and in patients with peripheral arterial disease, prior amputation, with end-stage renal disease (ESRD) and even on dialysis.

3.4.2. TWO₂ Only vs. Other Advanced Modalities (Direct Either/or Comparison)

A unique and clinically relevant aspect of the Yellin et al., analysis was a second comparison that directly evaluated TWO₂ only against other advanced therapies (NPWT, skin substitutes, growth factors).

Patients treated with TWO₂ alone experienced significantly better outcomes than those treated with only other advanced modalities, demonstrating an 88% relative reduction in hospitalization (6.9% vs. 58.8%, $p < 0.0001$) and 61% fewer amputations (13.8% vs. 35.3%, $p = 0.016$) at one year (Figure 2).

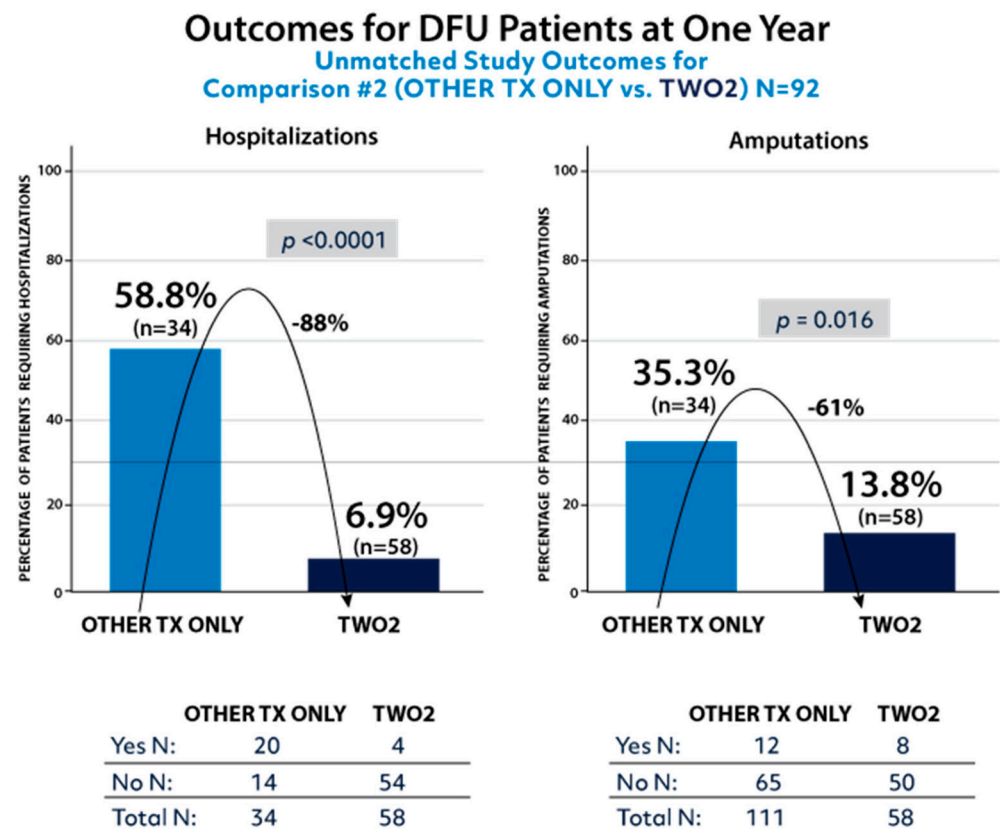


Figure 2. TWO₂ Only vs. Other Advanced Modalities: One Year Outcomes: Patients treated with TWO₂ alone experienced significantly better outcomes than those treated with only other advanced modalities, demonstrating an 88% relative reduction in hospitalization and 61% fewer amputations (Reprinted/adapted from ref. [30]).

These outcomes suggest that TWO₂ therapy not only offers additive benefit but may also be associated with significantly better outcomes compared with other traditionally used advanced wound care modalities when used as a primary adjunctive therapy. Prospective head-to-head randomized controlled trials remain needed to establish causality.

In sum, the study demonstrated that the use of TWO₂ therapy, with or without other adjunctive treatments, was associated with significantly reduced frequency of wound-related hospitalization and amputation for patients afflicted with DFUs. Given that both outcomes are associated with higher costs, increased healthcare utilization, and poorer quality of life, these findings highlight TWO₂ therapy’s potential value as a cost-effective, patient-centered modality in DFU management [2–4].

4. Discussion

4.1. Contextualizing TWO₂ Amongst Other Advanced Wound Technologies

The advanced wound care modalities in Table 1 each have defined indications and limitations relevant to chronic lower-extremity wound pathophysiology.

Table 1. Relative Comparison of Advanced Wound Therapies.

Therapy	pO ₂ Increase	Edema Reduction/Lymphatic Activation	Inflammation Resolution	Angiogenesis Upregulation	Bioburden Control	ECM/Cellular Repair	Unique Mechanism(s)
TWO ₂ [6,22]	+++	+++	+++	+++	++/+++	+++	Pressurized topical oxygen with non-contact cyclical compression; concurrent modulation of hypoxia, edema, and mechanotransduction signaling pathways
HBOT ** [31]	+++	+*	++	++	++	++	Systemic hyperoxygenation increasing dissolved plasma oxygen and tissue oxygen gradients
Continuous Delivery Oxygen [32,33]	++	–	++	++	++	++	Sustained low-flow topical oxygen diffusion maintaining continuous wound surface oxygen gradient
NPWT [34]	+*	+	++	++	+	++	Macro/micro deformation induced mechanotransduction with controlled exudate removal and wound edge stabilization
Skin Substitutes [35]	–	–	+	++	–	+++	Bioactive extracellular matrix scaffold providing structural support, cellular signaling, and growth factor modulation
Growth Factors [36]	–	–	+	++	–	++	Exogenous peptide-mediated activation of cell proliferation and angiogenic signaling pathways
Compression [37,38]	+*	+++	++	+	+	+	Venous hypertension reduction and lymphatic unloading through sustained, graduated external pressure

Relative Comparison of Advanced Wound Therapies: Ratings reflect the authors’ qualitative synthesis of dominant direct mechanisms reported in published mechanistic and clinical literature. “+++” denotes primary, robust engagement of the pathway supported by multiple published mechanistic and clinical studies; “++” denotes a consistent secondary contribution supported by published evidence; “+” denotes an indirect or context-dependent contribution; “–” denotes no established primary direct mechanism. Ratings are comparative synthesis tools and not absolute measures of clinical efficacy or relative effectiveness. * Indirect effects mediated via perfusion or edema modulation rather than direct oxygen delivery or antimicrobial action. ** Requires intact vasculature.

Hyperbaric oxygen therapy (HBOT) delivers 100% oxygen at 2.0 to 3.0 atmospheres absolute via systemic delivery, increasing dissolved plasma oxygen and tissue oxygen gradients. Cochrane evidence supports improved short-term healing in selected diabetic foot ulcers and refractory wounds [31]. Because delivery is systemic, efficacy depends on intact arterial inflow and microvascular perfusion, limiting use in severe peripheral arterial disease. Additional constraints include risks of barotrauma and oxygen toxicity, chamber availability, and patient suitability for hyperbaric exposure [31].

Continuous delivery topical oxygen (CDO) systems apply sustained low-flow, low-pressure oxygen (3 to 15 mL/h) under a sealed dressing. Controlled studies and a recent systematic review report improved healing in Wagner Grade 1 and 2 DFUs [32,33]. CDO does not reduce edema or restore lymphatic function, and the comparative evidence base remains smaller than for TWO₂ therapy [32,33].

Negative pressure wound therapy (NPWT) applies subatmospheric pressure via a sealed dressing, promoting granulation, exudate removal, and angiogenic signaling. It is best supported for wound bed preparation, exudate management, and acute traumatic or surgical wounds [34]. NPWT does not directly correct hypoxia or restore limb hemodynamics; practical considerations include clinical staff requirements for application (typically 2–3x/week), periwound maceration, treatment-related pain, and bleeding risk.

Bioengineered skin substitutes provide an exogenous extracellular matrix scaffold, with some formulations incorporating viable cellular components or growth-factor enriched constructs. FDA-approved living skin equivalents have demonstrated improved healing of DFUs and VLUs that have failed initial standard care [35]. Effectiveness requires a clean, well-perfused, well-oxygenated wound bed and is reduced in hypoxic or protease-rich environments. Multiple applications are often needed at substantial cost, with outcomes dependent on patient selection and wound bed preparation [35].

Growth factor therapies, such as recombinant human platelet-derived growth factor (becaplermin), deliver exogenous peptide signaling for fibroblast recruitment, angiogenesis, and granulation. Becaplermin is FDA-approved as an adjunct for neuropathic DFUs. Growth factor activity is reduced by proteolytic degradation in chronic wound exudate and by tissue hypoxia [36].

Compression therapy applies sustained, graduated external pressure, reducing venous hypertension and supporting lymphatic drainage. It is the standard of care for venous leg ulcers, with consistent evidence for improved healing and reduced recurrence [37,38]. Compression does not directly correct tissue hypoxia or provide cellular reparative signaling. Adherence is a recognized challenge, and use is limited or contraindicated in significant arterial insufficiency [37].

Across these modalities, therapies targeting a single component of chronic wound pathology may improve intermediate outcomes, whereas therapies addressing multiple interdependent drivers of chronicity may offer a more favorable mechanistic profile for durable healing, consistent with the comparative outcomes detailed in Section 3. A structured comparison is presented in Table 1.

4.2. Practical Considerations and Clinical Integration

TWO₂ therapy is designed for home-based administration by the patient, reducing treatment burden and improving adherence, while advancing access to care. It can be integrated into care pathways either as an adjunct to other advanced modalities or as an alternative when other therapies fail. Sequential strategies may include initial NPWT for exudate control followed by transition to TWO₂ for angiogenesis and epithelialization, or concurrent use with skin substitutes to optimize graft uptake.

4.3. Cost-Effectiveness Considerations

The following economic considerations are derived from retrospective cohort analyses and a published decision-analytic Markov model; they represent modeled estimates and should be interpreted in that context rather than as findings from prospective comparative cost analyses. The original authors did not conduct an independent health economic evaluation as part of this review.

Wound recurrence, hospitalizations, and amputations represent major cost drivers in chronic wound care. Repeat emergency department admissions, surgical intervention, rehabilitation, prosthetics, and long-term disability contribute to the economic burden. Even more difficult to quantify are the negative psychosocial and quality-of-life impacts on patients and caregivers.

The reductions in hospitalization (up to 88%) and amputation (up to 73%) observed in real-world TWO₂ cohorts suggest significant potential downstream cost savings. Additionally:

- Home-based therapy reduces outpatient visit frequency.
- Improved durability should reduce recurrence-related costs.
- Lower amputation rates should reduce lifetime disability expenditures.

A recent Markov model analysis suggested that TWO₂ may be cost-dominant compared with standard care, with modeled estimates indicating lower total two-year costs and improved quality-adjusted life years (QALYs). These projections are based on modeling assumptions and require confirmation through prospective economic evaluations comparing TWO₂ directly with NPWT, HBOT, and skin substitutes across longer time horizons [2].

4.4. Limitations

The evidence base for TWO₂ therapy includes both randomized and observational data, each with inherent limitations. Even though largely compensated for by robust study designs and statistical approaches such as propensity scoring, retrospective analyses may be subject to unmeasured confounding, while randomized trials may not fully capture real-world complexity. Direct head-to-head randomized comparisons with NPWT, HBOT, or bioengineered skin substitutes are currently lacking and represent a priority gap for future research. Additionally, the available comparative literature does not provide stratified efficacy analyses by wound chronicity, severity grade, or comorbidity burden, and the variable follow-up duration in the Lohr cohort (13.9 ± 4.9 months) limits direct interpretation of recurrence and hospitalization endpoints. The cost-effectiveness data summarized here derive from a published decision-analytic Markov model and do not constitute prospective economic evidence. Despite these limitations, the consistency of findings and long-term outcomes across study designs and wound types is supportive of the observed therapeutic benefits.

5. Conclusions

TWO₂ therapy is supported by a converging body of evidence demonstrating improved healing durability and substantial reductions in hospitalization and amputation in patients with multimorbid chronic lower-extremity wounds. The randomized TWO₂ Study establishes efficacy, while the real-world analysis by Yellin and colleagues demonstrates generalizability in far broader comorbid patient populations, as well as comparative effectiveness against other advanced wound modalities. The large cohort retrospective study by Lohr and colleagues further supports the generalizability of healing outcomes across varied lower extremity wound types, and in populations refractory to standard care and advanced therapies. By addressing hypoxia, edema, and impaired microcirculation simultaneously, TWO₂ therapy offers a mechanism-driven approach that translates into meaningful patient-centered outcomes [6,22,23,30].

These findings support the earlier and broader adoption of TWO₂ therapy in the management of chronic lower extremity wounds, with the potential to reduce wound recurrence, lower amputation risk, and decrease the economic burden and social toll of chronic wounds on patients, caregivers, and the healthcare system.

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