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Research Paper

Hyperbaric oxygen therapy in the healing process of foot ulcers in diabetic type 2 patients marked by interleukin 6, vascular endothelial growth factor, and PEDIS score: A randomized controlled trial study

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ABSTRACT

Introduction: Diabetic foot ulcers (DFU) is a major social issue in terms of treatment cost. DFU has a high risk of infection with morbidity and an increased risk of lower-extremity amputations. Currently, there are no satisfactory treatments for DFU. This motivates a search for methods that can stimulate the acceleration of wound healing; one of these methods is the use of hyperbaric oxygen therapy (HBOT). This study attempts to prove the ability of HBOT to accelerate the healing process of DFU by increasing levels of both serum Interleukin 6 (IL-6) and Vascular Endothelial Growth Factor (VEFG), assessed by the perfusion, extent, depth, infection and sensation (PEDIS) score.

Methods: Twenty DFU patients were divided into two groups. The control group underwent a holistic DFU treatment without HBOT and was tested twice for serum IL-6 and VEGF levels, 1 day apart. The HBOT group underwent a holistic DFU treatment with HBOT and was also tested twice for serum IL-6 and VEGF levels: one day before HBOT and 2 h after the first day of HBOT.

Results: The changes in serum IL-6 and VEGF levels were greater in patients with HBOT than in control patients ($p = 0.025$ and $p = 0.004$). As for PEDIS score assessment, the HBOT group had significantly lower PEDIS scores than the control group ($p < 0.001$).

Conclusion: HBOT can help accelerate the wound healing process, which was proven by increased serum IL-6 and VEGF levels and a lower PEDIS score.

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1. Introduction

Diabetes mellitus is an endocrine disorder that can cause many complications, one of which is diabetic foot ulcers (DFU). A DFU becomes a major social issue when it comes to the cost of treating it. DFU has a high risk of infection with a morbidity rate of 40–80%, and 14–20% of patients with DFU require amputation [1–4]. The rate of lower extremity amputations in diabetic patients is up to twenty times higher compared to those who are non-diabetic [5,6]. Around 70–80% of all non-traumatic amputations occur in patients with diabetes [7].

Currently, there are no satisfactory treatments for DFU [8]. This motivates the search for methods that can stimulate the acceleration of wound healing. One of these methods is hyperbaric oxygen therapy (HBOT) [9,10]. HBOT is a therapeutic method that uses 100% oxygen at pressures that are higher than the atmospheric pressure at sea level, which is at 2–3 atm absolute (ATA) in hyperbaric chambers [9–11].

Wound healing is a complex process that involves a series of overlapping phases: inflammation, epithelization, angiogenesis, and matrix deposition [12]. The process of wound healing involves interactions between oxidative stress and cytokine activity [13]. DFU patients who undergo additional HBOT are known to experience an increase in the process of angiogenesis, which results in faster reepithelization and granulation processes [14]. Kranke et al. identified a significant benefit of HBOT in reducing ulcer area at the end of a 30-day treatment for 30 patients with non-healing diabetic ulcers and venous ulcers (MD 61.88%, 95% CI 41.91 to 81.85, $p < 0.00001$) [15].

Interleukin 6 (IL-6) is a multifunctional cytokine that plays a key role in the immune system as well as in various biological processes, and it is a good regulator for both acute wounds and infections. It is a pleiotropic cytokine that has an important function in modulating collagen deposition and angiogenesis [16]. Similarly, Vascular Endothelial Growth Factor (VEGF)—a protein that promotes angiogenesis and is expressed in both endothelial and non-endothelial cells—is essential in the process of wound healing and restoring adequate blood circulation to the wound [17].

The aim of this study is to prove the ability of HBOT in accelerating wound healing in DFU patients through increased expressions of IL-6 and VEGF in the bloodstream, as well as a lower the perfusion, extent, depth, infection and sensation (PEDIS) score.

2. Methods

2.1. Study design and setting

This Randomized Controlled Trial (RCT) study was conducted from April through August 2019 in our institution. The HBOT groups and the control group were recruited from the same population of patients with diabetic foot ulcers who visited the surgical outpatient in our institution. The treatment group was randomly selected from patients who agreed to receive HBOT, whereas the control group comprised patients who opted to not receive the treatment. The protocol of the study was approved by the Institutional Review Board at our institution (no. 242/EC-KEPK/XII/2018) and has been registered with the Research Registry (no. 6217). This study has been reported in line with the Consolidated Standards of Reporting Trials (CONSORT) Guidelines [18,19].

2.2. Eligibility criteria

The inclusion criteria for this study were diabetic patients with DFU who have undergone or are undergoing holistic therapy for diabetes and who have an Ankle Brachial Index (ABI) of ≥0.9, a HbA1c value of ≤8%, an albumin serum of >3.5 g%, and a Hb value of ≥8 g%. Signed letters of consent were obtained for all included patients. The exclusion criteria consisted of type 1 diabetes, gestational diabetes, foot ulcers without a history of type 2 diabetes, coagulopathy, traumatic ulcers, malignancy, peripheral vascular disease, and conditions that contraindicate the use of HBOT.

2.3. HBOT procedure

HBOT was administered by the inhalation of 100% oxygen at a pressure of 2.4 ATA in a hyperbaric oxygen chamber for 60 min—which was divided into two 30-min periods, each with 10-min gaps. This treatment was received every day for 3 days.

2.4. IL-6 and VEGF sample examination

The blood samples (4–5 mL) were collected from a peripheral vein. The blood serum was separated in a plain vacuum tube, aliquoted and stored at −20 °C, and used for subsequent assays of IL-6 and VEGF in the Biomolecular and Immunology Laboratory in the Faculty of Medicine at Sam Ratulangi University in Manado, Indonesia. IL-6 and VEGF activity was measured spectrophotometrically in accordance with the kit protocol using a human IL-6 Quantikine ELISA (i.e., enzyme-linked immunosorbent assay) kit (catalog no. D6050) and a human VEGF Quantikine ELISA kit (catalog no. DVE00), both purchased from R&D Systems, Inc. (USA). Absorbance readings were obtained using an ELx800 microplate reader (Bio-Tek Instruments, Inc. USA) at a wavelength of 450 nm.

IL-6 and VEGF levels were measured twice in the control group: one day before and one day after receiving a holistic treatment for DFU. IL-6 and VEGF levels were measured twice in the HBOT group: one day before HBOT and 2 hours after the first session of HBOT. The control group received a PEDIS score assessment on the first and the third days, while the HBOT group was assessed before HBOT and after the third session of HBOT.

2.5. Statistical analysis

The statistical analysis was performed using R statistical software version 3.5.1 [20]. Descriptive tabulations were performed according to the type of variable. Numeric scale variables were presented as mean and standard deviation, or as median and interquartile range (IQR) if the Shapiro-Wilk test showed an abnormal distribution. Categorical variables are displayed in total and proportion.

3. Results

Twenty type 2 diabetes patients with DFUs were divided into two groups: a group who received HBOT and a control group. Each group consisted of ten patients. Patient characteristics can be seen in Table 1.

The change in the values of wound healing markers (delta), serum IL-6, VEGF, and PEDIS score are shown in Table 2. Overall, the patients in the study had elevated serum IL-6 levels, from a baseline median value of 8.1 pg/mL to a median value of 19.7 in the second measurement (one day after the clinical course began), for a median increase of 11.7 (IQR 10.5–16.4 pg/mL).

The HBOT group had a notably significant increase in IL-6 levels compared to the control group (median 14.9 pg/mL vs. 11.2 pg/mL, $p = 0.025$). IL-6 levels in the HBOT group were higher than in the control group at the baseline (median 12.1 pg/mL vs. 3.5 pg/mL, $p < 0.001$) and one day after the treatment (27.1 ± 14.5 pg/mL).

Table 1
Characteristics of type 2 diabetes patients.

Variable	Total (N = 20)		Therapy Groups				*Pa*
			HBOT (n = 10)		Control (n = 10)		
	μ±SD	Med (Q1; Q3)	μ±SD	Med (Q1; Q3)	μ±SD	Med (Q1; Q3)	
Gender, *n* (%)							
Female	6 (30)	*	2 (20)	*	4 (40)	*	0.628
Male	14 (70)	*	8 (80)	*	6 (60)	*	
Age (years)	56.7 ±9.5	*	54.8 ±9.7	*	58.6 ±9.4	*	0.385
Weight (kg)	*	59.0 (55.8; 64.2)	*	57.5 (56.2; 63.8)	*	60.0 (55.2; 65.2)	0.820
Height (cm)	163.8 ±7.8	*	163.2 ±7.8	*	164.5 ±8.1	*	0.708
BMI (kg/m^2)	*	22.5 (20.7; 23.5)	*	22.5 (21.4; 23.2)	*	21.8 (20.5; 23.9)	0.791
DM onset (years)	*	10.0 (5.0; 20.0)	*	16.5 (6.2; 20.0)	*	7.5 (5.0; 10.0)	0.088
Wound onset (weeks)	*	8.0 (4.0; 30.0)	*	30 (9.0; 48.0)	*	40 (4.0; 8.0)	0.023
Smokers, *n* (%)	4 (20)	*	3 (30)	*	1 (10)	*	0.582
Hemoglobin level (g/dL)	9.9 ± 1.6	*	9.8 ± 1.9	*	10.0 ± 1.4	*	0.774
Erythrocyte count (10^6/μL)	3.6 ± 0.6	*	3.4 ± 0.7	*	3.8 ± 0.5	*	0.228
Thrombocyte count (10^3/μL)	394.0 ± 114.3	*	424.0 ± 137.2	*	364.0 ± 820.3	*	0.251
HbA1c level (%)	6.4 ± 0.9	*	6.5 ± 0.9	*	6.3 ± 0.8	*	0.640
Albumin level	*	3.5 (3.5; 3.6)	*	3.5 (3.5; 3.6)	*	3.6 (3.5; 3.6)	0.730
ABI	*	0.9 (0.9; 1.0)	*	1.0 (0.9; 1.0)	*	0.9 (0.9; 1.0)	0.671

Notes: μ = average; SD = standard deviation; Med = median; Q1 = first quartile; Q3 = third quartile; BMI = body mass index; DM = diabetes mellitus; ABI = ankle brachial index; *Pa* = *t*-test.

Table 2
Changes in IL-6, VEGF, and PEDIS score.

Variable	Total (N = 20)		Therapy Groups				*Pa*
			HBOT (n = 10)		Control (n = 10)		
	μ±SD	Med (Q1; Q3)	μ±SD	Med (Q1; Q3)	μ±SD	Med (Q1; Q3)	
IL-6 (pg/mL)							
Baseline	*	8.1 (3.6; 11.9)	*	12.1 (11.1; 12.7)	*	3.5 (3.0; 4.8)	<0.001
Day 1	*	19.7 (14.8; 25.7)	*	27.1 (22.3; 34.0)	*	14.5 (13.3; 15.5)	<0.001
Delta	*	11.7 (10.5; 16.4)	*	14.9 (11.6; 20.4)	*	11.2 (9.5; 12.0)	0.025
VEGF (pg/mL)							
Baseline	15.9 ± 14.7	*	19.6 ± 3.3	*	12.2 ± 2.1	*	<0.001
Day 1	38.1 ± 12.9	*	47.8 ± 9.5	*	28.4 ± 7.1	*	<0.001
Delta	22.2 ± 9.9	*	28.2 ± 8.8	*	16.2 ± 7.1	*	0,004
PEDIS Score							
Baseline	*	5	*	4.2	*	6	0.412
Day 3	4.4 ± 1.3	*	3.9 ± 1.1	*	4.9 ± 1.3	*	0.078
Delta	*	−1	*	−2	*	0	0.001

Notes: μ = average; SD = standard deviation; Med = median; Q1 = first quartile; Q3 = third quartile; Pa = *t*-test or Mann-Whitney *U* test, according to normal distribution; HBOT = hyperbaric oxygen therapy.

VEGF levels in all patients showed an increase at the second measurement (one day after the HBOT was administered), with a mean increase of 22.2 ± 9.9 pg/mL from the baseline average of 15.9 ± 4.7 pg/mL. This increase was greater in the HBOT group than in the control group (mean 28.2 pg/mL vs. 16.2 pg/mL, $p = 0.004$).

The second PEDIS score assessment, which was taken on the third day after treatment, showed a decline compared to the baseline measurement. This decline was greater in the HBOT group than in the control group (−2 vs. 0, $p = 0.001$).

In Fig. 1, the HBOT's and control group's changes in IL-6 levels are compared between the baseline and the first day after treatment. It can be seen that HBOT group had higher IL-6 levels than the control group ($p = 0.025$).

Fig. 2 displays the changes in VEGF levels of the control group and of the HBOT group. It can be clearly seen that VEGF levels in the HBOT group were higher than in the control group ($p = 0.004$).

Fig. 3 shows that the decline in PEDIS score was more pronounced in the HBOT group than in the control group (p = 0.001). Lastly, Figs. 4–6 provide clinical evidence of improved wound appearance after the administration of HBOT.

4. Discussion

IL-6 and VEGF levels were significantly higher in the HBOT group than in the control group. Likewise, PEDIS scores after the third session of HBOT were notably lower in the HBOT group than in

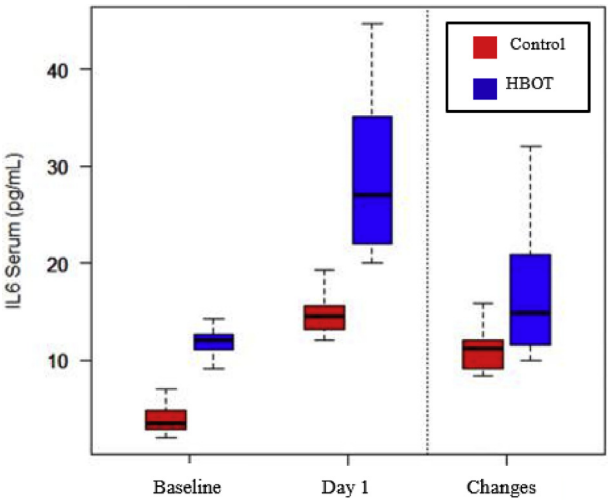

Fig. 1. Changes in IL-6 levels in the HBOT group vs. in the control group.

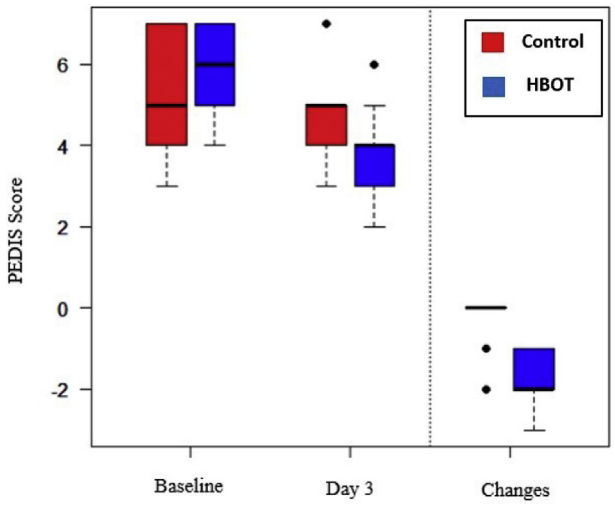

Fig. 3. Changes in PEDIS scores in the HBOT group vs. in the control group.

the control group. Both results suggest that HBOT can indeed accelerate fibroblast synthesis, accelerate collagen deposition, and stimulate macrophages in damaged tissue, thereby expediting the resolution of wound healing due to higher IL-6 and VEGF levels. At the start of the trial, the HBOT groups and the control group had different IL-6 and VEGF levels; the treatment group's were higher than the control group's. However, the differences at baseline were potentially compensated by remarkably high changes in IL-6 and VEGF levels in the HBOT group after treatment. Showing similar results, Sureda et al. [5] observed a significant increase in IL-6 levels among patients with chronic wounds after they received HBOT ($p < 0.05$). HBOT has also proven to be an effective adjuvant therapy in reducing ulcer size and improving wound outcomes [21,22].

In general, DFU treatments include standard care and adjuvant therapy. All patients in our study received not only standard care, comprising sharp debridement, dressing changes, blood glucose control, and vascular assessment, but also HBOT as an adjuvant therapy. Although other adjuvant therapies are currently available, including negative wound pressure therapy, treatment with acellular bioproducts, energy-based therapies, skin grafting, and bioengineered skin substitution [23], of those therapies our facility offers only negative wound pressure therapy, namely vacuum-assisted closure (VAC), which can indeed be more cost-effective than HBOT. However, we ultimately did not use VAC in view of our patients' multiple wound sites and because it requires hospital admission, whereas our patients were ambulatory. Although ambulatory patients are eligible to receive VAC, socioeconomic factors (e.g., level of education) often prevent families from reliably providing wound care at home. HBOT is used as a treatment option for DFU because it forces oxygen to diffuse intracellularly, thereby increasing the amount of oxygen in tissues [24,25]. High oxygen concentrations increase the production of ROS, which mediates the expression of key molecules in the inflammation, resolution, and wound repair processes. Therefore, the increase in the production of ROS is considered essential to how HBOT participates in the wound healing process [25–27].

The pleiotropic cytokine IL-6 has both pro-inflammatory and anti-inflammatory functions; IL-6 has been documented as both providing satisfactory wound healing results and helping in the inflammatory resolution phase. IL-6 increases the induction of

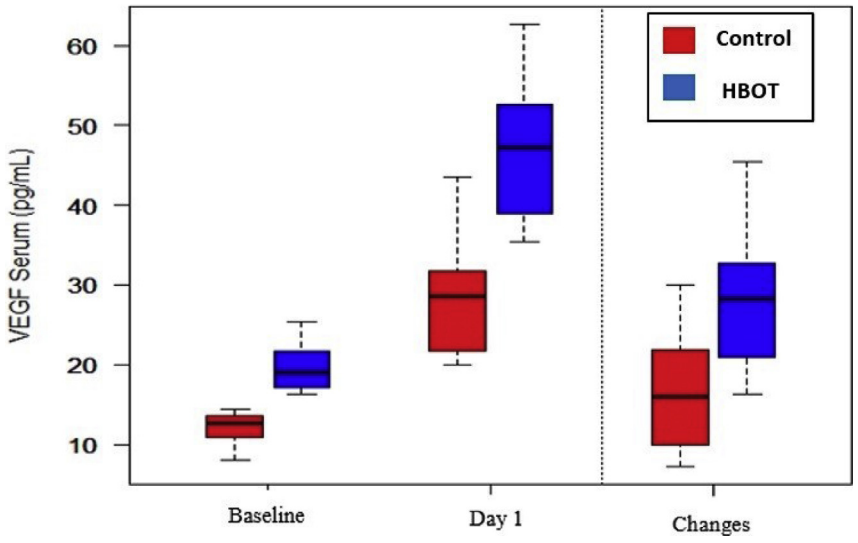

Fig. 2. Changes in VEGF levels in the HBOT group vs. in the control group.

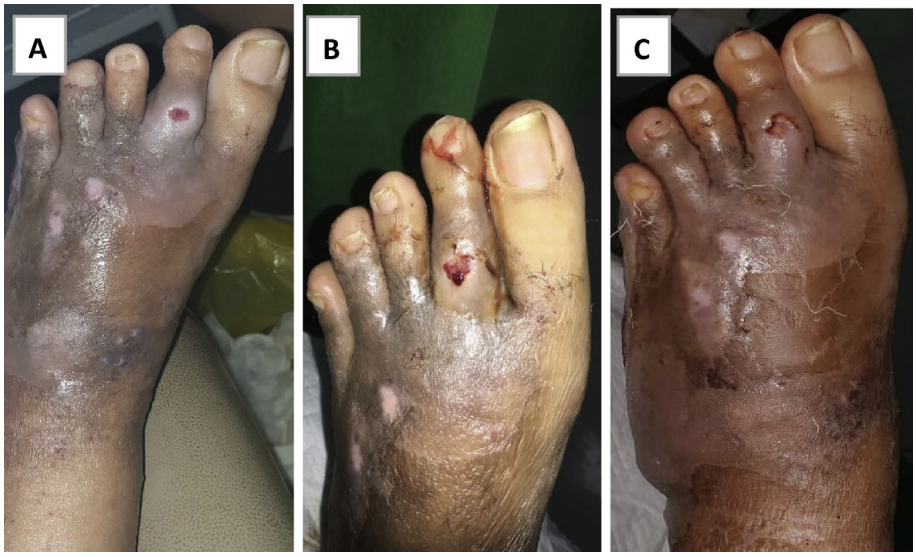

Fig. 4. A clinical photo of DFU from patient number 4: A) before HBOT; B) after the first session of HBOT; C) after the third session of HBOT, the wound appears moist and smaller in diameter.

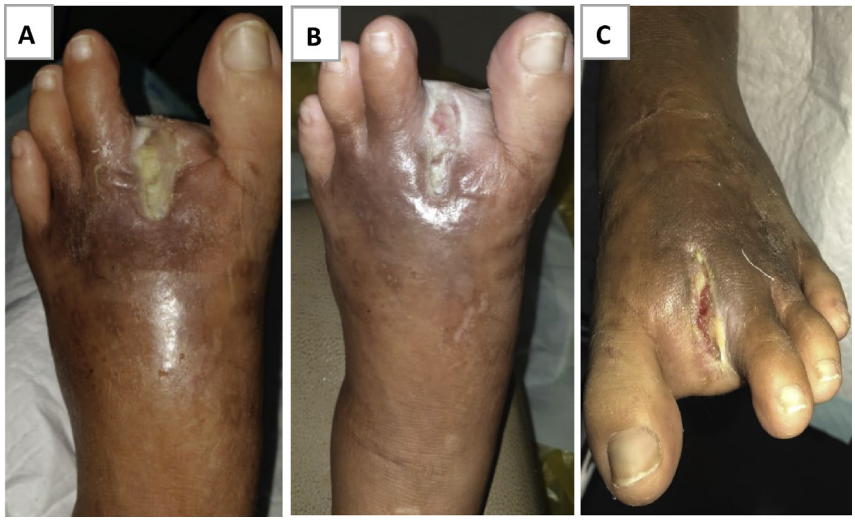

Fig. 5. A clinical photo of DFU from patient number 8: A) before HBOT, with a pale-looking wound; B) after the first session of HBOT; C) after the third session of HBOT, the wound appears smaller in diameter and granulating tissue has developed.

macrophages, which have anti-inflammatory and wound healing properties, through alternative pathways. The continuous IL-6 supply produced by frequent HBOT sessions is crucial in the wound healing process [27]. High oxygen concentration is needed in hypoxic wounds by the cells that participate in the process of wound healing (neutrophils, fibroblasts, and macrophages) to repair damaged tissues. This process of wound healing has proven that HBOT reduces the number of lower-limb amputation cases in diabetic patients.

An important intermediary factor in the processes of collagen deposition in wounds, cross-links, and neovascularization is the increased oxygen levels during HBOT [27,28]. Hao et al. studied the effects of HBOT on the expression of inflammatory mediators in keloid patients and found that levels of these mediators were significantly altered by HBOT administration, suggesting that HBOT is effective in reducing inflammatory responses and thus accelerating wound healing [28].

VEGF, a potent angiogenic factor, is an essential growth factor for vascular endothelial cells. VEGF is produced by many cell types, including macrophages, platelets, tumor cells, and more. VEGF plays a key role in normal physiological functions, such as bone formation, hematopoiesis, and—most importantly—wound healing [29]. VEGF production is subsequent to hypoxia. After an injury, the occurrence of hypoxia triggers hypoxia-inducible factor-1 (HIF-1), promoting angiogenesis by upregulating such target genes as VEGFA. The main isoform in the wound, VEGFA binds to its receptors on endothelial cells, fostering vessel growth [30]. This is why VEGFA is considered the most important positive regulator of angiogenesis. One study found that applying topical VEGFA to a diabetic wound in an animal model is able to accelerate wound healing [31].

The results of this study show a significant rise of VEGF levels in those who received HBOT compared to those who did not (mean 28.2 pg/mL vs. 16.2 pg/mL, $p = 0.004$). The notable difference

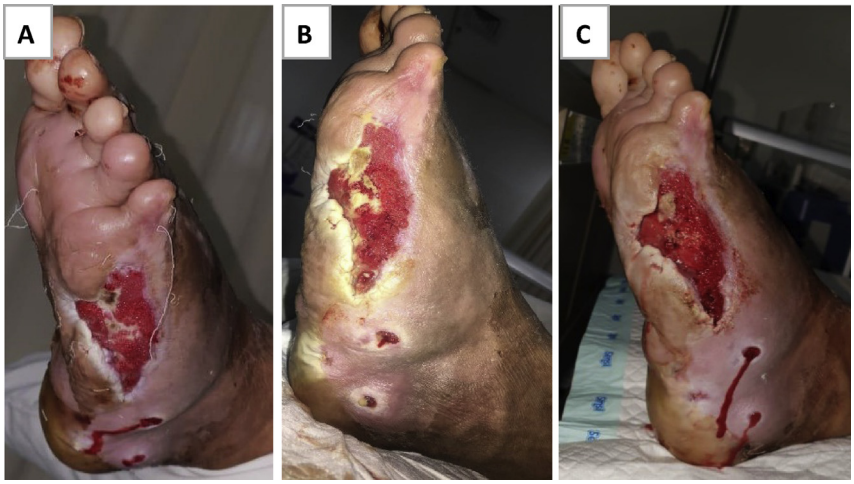

Fig. 6. A clinical photo of DFU from patient number 9: A) before HBOT, the wound edges appear pale and calluses are present nearby; B) after the first session of HBOT; C) after the third session of HBOT, a well-improved wound observed by its cherry-red, moist-looking appearance.

enables better interpretation of the VEGF measurement results in the two treatment groups both at the baseline and one day after the procedure. As observed, serum VEGF levels were generally higher in the HBOT group than in the control group at the baseline (19.6 pg/mL vs. 12.2 pg/mL, $p < 0.001$). Without a significant p value to mark the difference (VEGF delta) between the two groups, the VEGF measurements would have been difficult to interpret. Specifically, the higher VEGF levels in the HBOT group one day after the treatment (47.8 ± 28.4 pg/mL) could have been rejected as insignificant, as the VEGF levels in the HBOT group were already higher than in the control group at baseline. However, due to the significant change in delta VEGF values in HBOT patients, it is clear that this doubt can be eradicated.

Semadi [29] found a notable increase in the expression of VEGF after HBOT administration with humans after 2 weeks, and Susilo et al. [32] found the same after HBOT administration with animals for 5 days. This shows that HBOT can be an effective wound healing therapy for both humans and animals. Overall, these studies have shown that HBOT is capable of boosting the production of VEGF, resulting in an accelerated wound healing process in patients with DFU.

Although other blood markers were measured along with IL-6 and VEGF, we recorded only the levels of IL-6 and VEGF because they were relevant to the study. Other studies have also involved exploring different biomarkers with important roles in wound healing and that are affected by HBOT, including hypoxia-inducible factor 1 (HIF-1) [33], platelet-derived growth factor (PDGF), transforming growth factor beta (TGF-β), tumor necrosis factor alpha (TNF-α), preproendothelin 1 (PPET-1) and stromal cell derived factor 1 (SDF-1). At the same time, HBOT can reduce the levels of proinflammatory cytokines such as prostaglandin, IFN-γ, IL-1, and IL-10, which contributes to prolonged inflammation [34].

PEDIS scores were used to clinically assess DFUs as well as predict the outcomes of the ulcers. Although other methods of assessment are available, the PEDIS scoring system returns more accurate results than other methods such as the Wagner system and the Site, Ischemia, Neuropathy, Bacterial, Infection, and Depth (SINBAD) system [35]. By using objective techniques and comprehensive parameters in correspondence with ulcer healing, the PEDIS scoring system classifies DFUs into five categories—perfusion, wound area, wound depth, infection, and sensation—all based on the PEDIS classification system [35].

Perfusion is assessed through a combination of physical examination and non-invasive vascular testing. Clinical signs include the presence of dorsalis pedis artery pulsation or the posterior tibial artery of the affected foot. Non-invasive criteria include ankle-brachial index (ABI), toe-brachial index (TBI), transcutaneous oxygen pressure ($TcpO_2$), and ankle/toe pressure. The measurement of the wound area in cm^2 is calculated by multiplying the largest diameter and the second-largest diameter (measured perpendicular to the first diameter). The standard score in the wound area category is determined by assigning the ulcer to one of the following groups: intact skin, <1 cm^2, 1–3 cm^2, or >3 cm^2. The depth of the wound is evaluated using a sterile blunt nasal probe and an imaging test. Diagnosis of infection is based on signs of inflammation, presence of secretions, laboratory test results, and imaging tests. Sensation is evaluated using a 10-g monofilament and/or 128-Hz tuning fork on one or more legs [35]. Finally, all five categories of the PEDIS classification system are added up, resulting in an aggregate PEDIS score that ranges on a scale of 0–12. A PEDIS score of 7 or more is correlated with a higher likelihood of healing difficulties [35,36]; adverse outcomes, such as amputation and unhealed ulcers, are encountered in patients with high PEDIS scores [36].

The PEDIS scoring system is able to quickly and accurately assess the development of wound healing in DFUs, as it is considered the best method to predict wound outcomes compared to other scoring systems [35,37]. A study by Li et al. [38] shows that a PEDIS score of 7 has a greater risk for developing adverse outcomes. In predicting adverse outcomes, the PEDIS score has a sensitivity of 93% and a specificity of 82%. These values are preferred over the SINBAD and Wagner scoring systems, which have 90% and 88% sensitivity, respectively, and 73% and 80% specificity, respectively [22,38]. A limitation of the study, however, was that other biomarkers affected by HBOT that aid wound healing were not measured due to the lack of resources at our facility. Further research could be performed to explore other biomarkers affected by HBOT.

5. Conclusion

The results of this study revealed that HBOT can accelerate the wound healing process in patients with DFU. This is closely related to the effects of HBOT on increasing the levels of both serum IL-6 and VEGF, which is further confirmed by a lower PEDIS score. A

future study needs to be conducted on the effects of HBOT in the process of wound healing, in order to explore future applications of this therapy.

Ethical approval

All procedure for human experiment has been approved by the Institutional Review Board of our institution number: 242/EC-KEPK/XII/2018.

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Author contribution

MHO, MCO, DET, SWS, MH, AAI, and ERS wrote the manuscript and participated in the study design., MHO, MCO, DET, SWS, ERS, DMA, and JAK drafted and revised the manuscript. MHO, MCO, DET, SWS, ERS, DMA, and JAK, performed treatment and surgery. MHO, MCO, DET, SWS, and ERS performed bioinformatics analyses and revised the manuscript. All authors read and approved the final manuscript.

Conflict of interest statement

The authors declare that they have no conflict of interests.

Guarantor

Mendy Hatibie Oley.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ijso.2020.11.012.

References

[1] Pemayun TGD, Naibaho RM, Novitasari D, Amin N, Minuljo TT. Risk factors for lower extremity amputation in patients with diabetic foot ulcers: a hospital-based case-control study. Diabet Foot Ankle 2015;6:29629. https://doi.org/10.3402/dfa.v6.29629.
[2] Frykberg RG, Zgonis T, Armstrong DG, Driver VR, Giurini JM, Kravitz SR, et al. Diabetic foot disorders. A clinical practice guideline (2006 revision). J. Foot Ankle Surg. Off. Publ. Am. Coll. Foot Ankle Surg. 2006;45:S1–66. https://doi.org/10.1016/S1067-2516(07)60001-5.
[3] Chidiac C, Bru J-P, Choutet P, Decazes J-M, Dubreuil L, Leport C, et al. Management of diabetic foot infections. Med Maladies Infect 2007;37:14–25. https://doi.org/10.1016/j.medmal.2006.10.001.
[4] Yasa KP, Siki-Kawiyana K, Budhiartha AAG. Debridement with fasciotomy on diabetic foot reduces Tumor Necrosis Factor Alpha and increases Vascular Endothelial Growth Factor plasma with clinical improvement. Bali Med J 2014;3:97–103. https://doi.org/10.15562/bmj.v3i2.82.
[5] Sureda A, Batle JM, Martorell M, Capó X, Tejada S, Tur JA, et al. Antioxidant response of chronic wounds to hyperbaric oxygen therapy. PloS One 2016;11:e0163371. https://doi.org/10.1371/journal.pone.0163371.
[6] Organization WH. Global report on diabetes. World Health Organization; 2016.
[7] Armstrong DG, Boulton AJM, Bus SA. Diabetic foot ulcers and their recurrence. N Engl J Med 2017;376:2367–75. https://doi.org/10.1056/NEJMra1615439.
[8] Kessler L, Bilbault P, Ortéga F, Grasso C, Passemard R, Stephan D, et al. Hyperbaric oxygenation accelerates the healing rate of nonischemic chronic diabetic foot ulcers: a prospective randomized study. Diabetes Care 2003;26:2378–82. https://doi.org/10.2337/diacare.26.8.2378.
[9] Flood M. Hyperbaric oxygen therapy for diabetic foot ulcers. J. Lancaster Gen. Hosp. @BULLET Winter 2007;2.
[10] Bhutani S, Vishwanath G. Hyperbaric oxygen and wound healing. Indian J Plast Surg 2012;45:316–24. https://doi.org/10.4103/0970-0358.101309.
[11] Sahni T, Hukku S, Jain M, Prasad A, Prasad R, Singh K. Recent advances in hyperbaric oxygen therapy, vol. 14. Assoc Physicians India; 2003.
[12] Goldstein IM, Snyderman R, Gallin JI. Inflammation: basic principles and clinical correlates. Raven Press; 1992.
[13] Halliwell B, Gutteridge JM, Cross CE. Free radicals, antioxidants, and human disease: where are we now? J Lab Clin Med 1992;119:598–620.
[14] Hanft JR, Pollak RA, Barbul A, van Gils C, Kwon PS, Gray SM, et al. Phase I trial on the safety of topical rhVEGF on chronic neuropathic diabetic foot ulcers. J Wound Care 2008;17(30–32):34–7. https://doi.org/10.12968/jowc.2008.17.1.27917.
[15] Kranke P, Bennett MH, Martyn-St James M, Schnabel A, Debus SE, Weibel S. Hyperbaric oxygen therapy for chronic wounds. Cochrane Database Syst Rev 2015;2015:CD004123. https://doi.org/10.1002/14651858.CD004123.pub4.
[16] Tian Y-W, Stacey MC. Cytokines and growth factors in keratinocytes and sweat glands in chronic venous leg ulcers. An immunohistochemical study. Wound Repair Regen 2003;11:316–25. https://doi.org/10.1046/j.1524-475X.2003.11502.x.
[17] Pandey AK, Singhi EK, Arroyo JP, Ikizler TA, Gould ER, Brown J, et al. Mechanisms of VEGF (vascular endothelial growth factor) inhibitor-associated hypertension and vascular disease. Hypertension (Dallas) 2018;71:e1–8. https://doi.org/10.1161/HYPERTENSIONAHA.117.10271.
[18] Agha RA, Borrelli MR, Vella-Baldacchino M, Thavayogan R, Orgill DP, Pagano D, et al. The STROCSS statement: strengthening the reporting of cohort studies in surgery. Int J Surg 2017;46:198–202. https://doi.org/10.1016/j.ijsu.2017.08.586.
[19] Schulz KF, Altman DG, Moher D. CONSORT 2010 Statement: updated guidelines for reporting parallel group randomised trials. BMJ 2010;340:c332. https://doi.org/10.1136/bmj.c332. c332.
[20] R Core Team, Language R:A. And environment for statistical computing. 2018. https://www.r-project.org/.
[21] Ueno T, Omi T, Uchida E, Yokota H, Kawana S. Evaluation of hyperbaric oxygen therapy for chronic wounds. J Nippon Med Sch 2014;81:4–11. https://doi.org/10.1272/jnms.81.4.
[22] Stoekenbroek RM, Santema TB, Legemate DA, Ubbink DT, van den Brink A, Koelemay MJW. Hyperbaric oxygen for the treatment of diabetic foot ulcers: a systematic review. Eur. J. Vasc. Endovasc. Surg. Off. J. Eur. Soc. Vasc. Surg. 2014;47:647–55. https://doi.org/10.1016/j.ejvs.2014.03.005.
[23] Everett E, Mathioudakis N. Update on management of diabetic foot ulcers. Ann N Y Acad Sci 2018;1411:153–65. https://doi.org/10.1111/nyas.13569.
[24] Ontario HQ. Hyperbaric oxygen therapy for the treatment of diabetic foot ulcers: a health technology assessment, ont. Health Technol. Assess. Ser. 2017;17:1–142. https://pubmed.ncbi.nlm.nih.gov/28572866.
[25] Thackham JA, McElwain DLS, Long RJ. The use of hyperbaric oxygen therapy to treat chronic wounds: a review. Wound Repair Regen. Off. Publ. Wound Heal. Soc. [and] Eur. Tissue Repair Soc. 2008;16:321–30. https://doi.org/10.1111/j.1524-475X.2008.00372.x.
[26] Lipsky BA, Berendt AR. Hyperbaric oxygen therapy for diabetic foot wounds: has hope hurdled hype? Diabetes Care 2010;33:1143–5. https://doi.org/10.2337/dc10-0393.
[27] Sallam AAW, El-Sharawy A. Role of interleukin-6 (IL-6) and indicators of inflammation in the pathogenesis of diabetic foot ulcers. Aust. J. Basic Appl. Sci. 2012;6:430–5.
[28] Hao Y, Dong X, Zhang M, Liu H, Zhu L, Wang Y. Effects of hyperbaric oxygen therapy on the expression levels of the inflammatory factors interleukin-12p40, macrophage inflammatory protein-1β, platelet-derived growth factor-BB, and interleukin-1 receptor antagonist in keloids. Medicine (Baltim) 2020;99:e19857. https://doi.org/10.1097/MD.0000000000019857. e19857.
[29] Semadi NI. The role of VEGF and TNF-alpha on epithelialization of diabetic foot ulcers after hyperbaric oxygen therapy., open access maced. J Med Sci 2019;7:3177–83. https://doi.org/10.3889/oamjms.2019.297.
[30] Okonkwo UA, DiPietro LA. Diabetes and wound angiogenesis. Int J Mol Sci 2017;18. https://doi.org/10.3390/ijms18071419.
[31] Gurtner GC, Werner S, Barrandon Y, Longaker MT. Wound repair and regeneration. Nature 2008;453:314–21. https://doi.org/10.1038/nature07039.
[32] Susilo I, Devi A, Purwandhono A, Warsito SH. Effects of hyperbaric oxygen therapy in enhancing expressions of e-NOS, TNF-α and VEGF in wound healing. J. Phys. Conf. Ser. 2017;853:12030. https://doi.org/10.1088/1742-6596/853/1/012030.

[33] Sunkari VG, Lind F, Botusan IR, Kashif A, Liu Z-J, Ylä-Herttuala S, et al. Hyperbaric oxygen therapy activates hypoxia-inducible factor 1 (HIF-1), which contributes to improved wound healing in diabetic mice. Wound Repair Regen 2015;23:98–103. https://doi.org/10.1111/wrr.12253.

[34] Johnston BR, Ha AY, Brea B, Liu PY. The mechanism of hyperbaric oxygen therapy in the treatment of chronic wounds and diabetic foot ulcers. R I Med J 2013;99 (2016) 26–9, http://www.ncbi.nlm.nih.gov/pubmed/26827082.

[35] Chuan F, Tang K, Jiang P, Zhou B, He X. Reliability and validity of the perfusion, extent, depth, infection and sensation (PEDIS) classification system and score in patients with diabetic foot ulcer. PloS One 2015;10:e0124739. https://doi.org/10.1371/journal.pone.0124739.

[36] Gandhi C, Kadam P, Kamepalli V, Kadam Y. PEDIS grading and its role in diabetic foot ulcer management. Int Surg J 2019;6:2548–52. https://doi.org/10.18203/2349-2902.isj20192990.

[37] Monteiro-Soares M, Boyko EJ, Jeffcoate W, Mills JL, Russell D, Morbach S, et al. Diabetic foot ulcer classifications: a critical review. Diabetes. Metab. Res. Rev. 2020;36:e3272. https://doi.org/10.1002/dmrr.3272.

[38] Li N, Meng X-E, Guo D-Z, Fan D-F, Pan S-Y. Wound healing process and related laboratory indexes in patients with type 2 diabetes mellitus after hyperbaric oxygen intervention. Biomed Res 2017;28:8838–43.