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SYSTEMATIC REVIEW

EVALUATION OF THE EFFECTIVENESS OF COMBINATION ULTRASOUND TREATMENT AND BASIC CARE IN THE HEALING OF PRESSURE ULCERS: A SYSTEMATIC REVIEW

Christina Sotnikova¹, Polyxeni Bibou², Georgia Fasoi³, Georgia Toyli⁴, Martha Kelesi⁵

1. RN, MSc, Department of Anesthesiology, General Hospital of Athens "Korgialenio-Benakeio", Hellenic Red Cross, Greece
2. Lecturer, Nursing Department, University of West Attica, Greece
3. Professor, Nursing Department, University of West Attica, Greece
4. Assistant Professor, Nursing Department, University of West Attica, Greece
5. Professor, Nursing Department, University of West Attica, Greece

Abstract

Introduction: When the pressure ulcer does not respond to established standard care, then international guidelines recommend the use of alternative forms of therapy such as electrical stimulation, negative pressure wound treatment, ultraviolet radiation, electromagnetic field therapy or ultrasound. Low and high frequency ultrasound is used to treat various types of chronic ulcers including venous, diabetic and pressure ulcers.

Aim: The aim of the present systematic review was to investigate the effectiveness of ultrasound in promoting the healing of pressure ulcers.

Method and Material: A systematic review was carried out in the electronic databases PubMed and Cochrane Library. Inclusion criteria were primary and secondary studies, whose sample were patients with pressure ulcers, regardless of age, published in English language of the last decade (2008-2018).

Results: The review revealed 5 articles that met the inclusion criteria. The results of these studies showed an advantage in the use of ultrasound for the treatment of pressure ulcers in relation to the standard wound care only.

Conclusions: The use of ultrasound seems to be a process which improves and accelerates the healing of pressure ulcers. The method of low and high frequency ultrasound in the treatment of chronic wounds, presents many advantages. It appears to be easy to use, painless, less stressful and better accepted by patients.

Keywords: Ultrasound therapy, ultrasound treatment, pressure ulcers, wound healing.

Corresponding Author: Christina Sotnikova, Tel: 6938953755, E-mail: chris_x30@yahoo.com

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INTRODUCTION

Pressure ulcers (PrUs) represent a significant health challenge with a great number of people affected worldwide. Diseases that lead to restricted mobility and affect the blood and oxygen delivery to tissues play an especially important role in the development of PrUs. External factors such as pressure, friction, shear and moisture, as well as endogenous factors such as malnutrition, advanced age, hypotension, emotional stress, smoking and skin temperature, also play a combined role in the development of PrUs.¹

When the pressure ulcer does not respond to standard wound care, then the international guidelines recommend the use of alternative forms of treatment such as electrical stimulation (ES), negative pressure wound treatment (NPWT), ultraviolet radiation, pulsed electromagnetic field therapy (PEMF) or ultrasound (US).²⁻⁷ Ultrasound has been used by several health professionals for over 60 years. The biological effects on tissues exposed to ultrasound were first reported by Wood and Loomis in 1927.⁸ The application of ultrasound for therapeutic purposes began in Germany in the late 1930s and in the United States in early 1940s.⁹

Studies of the last decade have shown that ultrasound treatment has a greater effect on wound healing than standard wound care only.^{1,10-13}

The mechanism of action of ultrasound treatment

The therapeutic effects of ultrasound have been used by several researchers in the past. In 1960 Paul et al.,¹⁴ describe the treatment of 23 patients with pressure ulcers using ultrasound. This study concluded that it would be extremely useful to further investigate this specific treatment. In 1985, 25 years later, McDiarmid et al.¹⁵ published the first randomized clinical trial on the effects of ultrasound in healing of pressure ulcers. Although McDiarmid et al., could not show overall beneficial effect, they found that ultrasound treatment appeared to enhance the healing rate in a group of patients with infected surface ulcers.¹⁶ In a randomized clinical study of Nussbaum et al., 1994¹⁷ conducted in Canada, ultrasound treatment appeared Sotnikova et al.

to have a greater effect on wound healing than standard wound care alone.

Types of ultrasound

According to Kloth and Niezgodna,⁹ mechanical forces, such as ultrasound (US), can stimulate signal transduction pathways in tissue and produce a wide range of cellular effects that improve the healing of wounds.¹³

The ulcers can be managed with Low-Frequency Ultrasound (LFUS 22.5–40 kHz) or High-Frequency Ultrasound (HFUS; 1–3 MHz). The HFUS stimulates the conductivity of the cell membrane and increases the cell calcium concentration, which may promote significantly the activity of cells that are important for the wound healing. Moreover, the HFUS can stimulate the activity of macrophages and fibroblasts and activate the synthesis of collagen. In addition to this, the HFUS activates growth factors in wounds and promote neoangiogenesis.^{9,12,13}

The Low Frequency Ultrasound has been used for the treatment of various types of chronic wounds including pressure ulcers.^{10,18,19} The LFUS wound debridement offers a less traumatic, less painful surgical debridement, achieving destruction of the bacterial biomembrane, removing necrotic tissues, fibrosis, exudate and accelerating the formation of granulation tissue.²⁰⁻²² The LFUS waves act through two mechanisms: micro-cavitation and acoustic flow. Cavitation leads to cellular changes, destruction of periwound tissue in the ultrasound wave, also causes a rapid lysis of the necrotic tissue and ulceration fibrosis. The acoustic flow increases cell permeability and activates the intracellular secondary transport system which in turn causes an increase in collagen synthesis and production, growth factors, increase angiogenesis, macrophage response and fibrinolysis.^{9,10,18,22}

AIM

The aim of this systematic review was to investigate the effectiveness of ultrasound in promoting the healing of pressure ulcers.

METHODOLOGY

Articles for this systematic review were searched in PubMed and Cochrane Library databases, with the keywords ultrasound therapy, ultrasound treatment, pressure ulcers, wound healing. Only Clinical Trials, Controlled Clinical Trials, Meta-Analysis, Multicenter Studies, Randomized Controlled Trials, Systematic Reviews, published in the last 10 years (2008-2018) were included.

Inclusion Criteria:

- Studies in English language
- Published in the last 10 years (2008-2018)
- Studies of patients of all ages

Exclusion criteria:

- To refer to animals
- Other language

Search results were: PubMed 23 studies of which 2 were Controlled Clinical Trials, 3 Multicenter Study, 8 Randomized Controlled Trial and 10 Systematic Reviews. In Cochrane Library found 24 studies, 2 Controlled Clinical Trials, 4 Multicenter Studies, 15 Randomized Controlled Trials, 3 Systematic Reviews. The 8 duplicates were excluded due to the title as well as 15 articles published before 2008. The 24 articles were remained. Of the 24 articles, 15 were excluded due to the title (venous ulcers, diabetic foot). Articles obtained for further evaluation were 9. There were excluded 4 articles due to non-relevance to the subject. Finally 5 articles were remained for further research (flow chart). The description of these studies is presented in the table below.

RESULTS

In a prospective randomized study by Bora et al.¹, 27 treated neurological patients with stage II - IV pressure ulcers were studied using High Voltage Electrostimulation (15 patients) and Ultrasound (12 patients) for a period of 12 weeks. The researchers compared the effectiveness of these two methods in the treatment of pressure ulcers. A statistically significant reduction in pressure ulcer was found in both treatment methods Sotnikova et al.

with $p < 0.001$. However, the limitations of this study indicate that there was no homogeneity in the sample in terms of classification and ulcer surface due to randomization, plus the absence of the control group.

In a prospective randomized single blind study by Polak et al.¹³, the effectiveness between High Frequency Ultrasound and Electrostimulation was evaluated. The 77 patients with 88 stages II - IV pressure ulcers were studied. Patients were divided into 3 groups. The first group (28 patients) received the standard wound care only, the second group (25 patients) received the standard wound care with simultaneous use of ultrasound and the third group (24 patients) received the standard wound care with the electrostimulation. The results showed significant healing rates in all three groups, but the groups of methods tested had a statistically significant reduction in the surface area of pressure ulcers. A significant limitation was the short duration of monitoring the development of ulcers (4-6 weeks), the long duration of the study and especially the small sample of patients.

In another perspective, randomized, controlled clinical trial by Polak et al.¹², the effectiveness of High Frequency Ultrasound in stage II and III pressure ulcers in combination with standard wound care versus standard wound care only was evaluated and compared. The 42 patients with 44 pressure ulcers were studied. The patients were divided into one group which received a standard wound care only (22 patients with 23 pressure ulcers) and a second group which received a standard wound care in combination with ultrasound (20 patients with 21 pressure ulcers). The duration of follow-up was 6 weeks. The combination of the standard wound care with the ultrasound showed statistically significant results in terms of the average reduction of the ulcer area, on a weekly basis, over a period of 6 weeks and on healing in 6 weeks. In addition, the combination of the standard wound care with the use of ultrasound appeared to be superior although there were no statistically significant results. The limitations of the study includes the lack of a control group (sham ultrasound), that the study was not

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blinded as well as the researchers' belief that the standard wound care of pressure ulcers varied between the four research centers that participated in the study.

In a pilot randomized controlled clinical trial of Maeshige et al.¹¹, evaluated the effect of ultrasound compared with the standard wound care in the treatment of pressure ulcers. There were 5 elderly neurological patients, with 7 stage III and IV pressure ulcers, without local wound infection and extensive necrotic tissue as well as these patients did not have diabetes mellitus type 2 or peripheral arterial disease. The ultrasound treatment group consisted of 3 patients with 4 ulcers and the control group consisted of 2 patients with 3 ulcers. All patients underwent the standard wound care during the study. In period A, each pressure ulcer received the standard wound care only for 2-4 weeks. In period B, each pressure ulcer received the standard wound care in combination with ultrasound or the standard wound care with sham ultrasound for the same duration as an ABABA sequence (alternating periods A, B, A). The results showed superiority in the ultrasound treatment group in the specific time period compared to the control group. The authors cite as a limitation of the study the very small sample, although there is a predominant tendency in favor of combination treatment.

A prospective experimental study by Serena et al.¹⁰, assessed the effectiveness of non-touch low frequency ultrasound treatment (MIST Therapy) in reducing the bacterial colonization in stage III pressure ulcers and promoting the healing. This was a multicenter study with a total sample of 11 patients with chronic stage III pressure ulcers, who met criteria such as: wound volume up to 160 cm³, bacterial measurements > 105 CFU/g of tissue (without infection) as well as patients without clinical signs of acute ulcer infection, without head or neck injury and without malignancy in the wound bed. The bacterial load of the ulcer was measured before and 2 weeks after the MIST Therapy. All patients underwent the standard wound care during the study. No local antibiotics, antiseptics, analgesics and antimicrobial dressings were allowed. The results showed

that the bacterial load was significantly reduced with the MIST treatment. The *Streptococcus G.* showed different response and resistance. The authors cited the absence of a control group as a limitation of the study, although the non-touch low frequency ultrasound treatment was shown to reduce the bacterial load in the ulcer bed promoting the healing.

DISCUSSION

The results of this systematic review showed superiority in the use of ultrasound for the treatment of pressure ulcers in relation to the daily standard wound care. Different types of ultrasound are used for the same expected result. In particular, the high frequency ultrasound (HFUS) acts at the molecular level by stimulating a number of mechanisms that promote the healing process.^{1,11-13} On the other hand, the low frequency ultrasound (LFUS) is mainly used for the non-traumatic removal of soft necrotic tissue and biofilm. The presence of the biofilm reduces the effectiveness of the systematic use of antibiotics. Furthermore, chronic ulcers due to microangiopathy and fibrosis prevent the absorption of local antimicrobial agents, thus creating the need for non-toxic methods of cleaning such wounds. Regardless of the type of ultrasound, it appears that this method improves and accelerates the healing.^{10,13,18}

Of course, the studies have several methodological problems, which require the adoption of their results with caution and skepticism. A major problem is the small sample of patients used in each study as well as the lack of a common methodology and heterogeneity of ultrasound types.^{1,10-13}

In the study by Polak et al.¹², a total of 4 centers for elderly patients with pressure ulcers were used, where, as the authors emphasize, there is a possibility of different protocols in the daily standard wound care. In the studies of Bora et al.¹ and Serena et al.¹⁰ there were no control group.

Due to different methodology, follow-up period, use of different types of ultrasound and small sample, one can not draw safe conclusions in favor of the use of ultrasound.

LIMITATIONS

The limitations of this systematic review are the small number of studies, the search of the relevant bibliography in only two electronic databases and the fact that were used studies published in English language only. Another limitation is that the studies have been found that examine in addition to ultrasound and another method of therapeutic intervention such as electrostimulation. However, these studies gave superiority of ultrasound over the electrostimulation.^{1,13}

CONCLUSIONS

In conclusion, the use of ultrasound in the treatment of pressure ulcers appears to promote healing of the ulcer. However, the existence of a few studies with a limited sample can not lead to a generalization of the findings. For this reason, it is proposed to make more primary and multicenter studies on the use of ultrasound in combination with the daily standard wound care that might enable safer conclusions. Searching in more electronic databases as well as studies in a language other than English, would probably yield a larger volume of studies on this topic.

Taking into account the results of relevant researches, the method of low and high frequency ultrasound in the treatment of chronic wounds, presents many advantages. It seems to be easy to use, painless, less stressful and better accepted by patients.

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ANNEX

Flow Chart 1. Flow chart illustrating systematic review process.

TABLE 1. The results of the systematic review on the effect of ultrasound on the healing of pressure ulcers.

AUTHOR-YEAR-COUNTRY	AIM	TYPE OF STUDY-SAMPLE	DATA COLLECTION	ANALYSIS	RESULTS	LIMITATIONS-CONCLUSIONS
1. Bora et al., 2017, Turkey.	Assessment and comparison of the effectiveness of High Voltage Electrical Stimulation (HVES) with ultrasound (US), in treating Stage II-IV pressure ulcers of patients admitted to a rehabilitation center.	<p>Prospective, controlled trial.</p> <p>A total of patients (n=27) with pressure ulcers Stage II-IV (n=47).</p> <p>Stage II: 14 (29,8%)</p> <p>Stage III: 26 (55,3%)</p> <p>Stage IV: 7 (14,9%)</p> <p>Patients in the US group (n=12)</p> <p>Patients in the HVES group (n=15)</p> <p>Study duration: not mentioned</p>	<p>The patients' PrUs were classified according to the staging system developed by the National Pressure Ulcer Advisory Panel (NPUAP)</p> <p>HVES device: session duration was 60', 3 times/week for 4-12 weeks.</p> <p>US device: 3 times per week for 4-12 weeks.</p> <p>All PrUs (single or multiple) were photographed and dimensions were calculated before and after treatment using wound evaluation scales.</p>	<p>Data analysis: SPSS version 15.0.</p> <p>The normality of distribution: Shapiro-Wilk test.</p> <p>Comparison of repetitive measurements within the groups: Paired-sample t test.</p> <p>Non-parametric data with non normal distribution: Wilcoxon signed rank test.</p> <p>Nominal variables: Pearson χ^2 test.</p> <p>Intragroup correlations between variables: Spearman correlation test.</p> <p>Multivariate linear regression analysis was performed for variables affecting treatment outcomes.</p> <p>p < 0,05 was considered statistically significant.</p>	<p>Statistically significant reduction in pressure ulcer in both groups (43% in the HVES group and 63% in the US group (p < 0.001).</p> <p>HVES group: Stage II: 17,40 → 11,40 (p=0,012) Stage III: 28,53 → 20,66 (p<0,001) Stage IV: 30,78 → 18,64 (p=0,020)</p> <p>US group: Stage II: 7,61 → 1,50 (p=0,041) Stage III: 20,19 → 11,24 (p<0,001) Stage IV: -</p>	<p>Inability to provide intergroup homogeneity in terms of Pressure Ulcer Stage and Wound Surface Area because of randomization.</p> <p>Absence of the control group (Standard Wound Care-SWC)</p> <p>Both methods promise good results in reducing the traumatic surface and healing pressure ulcers.</p>
2. Polak et al., 2016,	Evaluation and comparison of effi-	Randomized, single-blind, controlled trial.	Prognosis for pressure ulcers with Nor-	Patients' characteristics were analyzed	Reduction in the surface area of PUs	Small number of qualifying patients, long

Poland.	ciency between high frequency ultrasound (HFUS) and electrical stimulation (ES).	<p>Hospitalized patients in 2 types of nursing and care centers—residential care facilities and temporary care facilities (n=77), with pressure ulcers (n=88).</p> <p>Stage II: 60 (68.1%)</p> <p>Stage III: 22 (25.0%)</p> <p>Stage IV: 6 (6.8%)</p> <p>Patient group with Standard Wound Care (SWC) (n=28)</p> <p>Patient group with SWC+US (n=25)</p> <p>Patient group SWC+ES (n=24)</p> <p>Study duration: 3 years.</p>	<p>ton scale, Braden scale, nutritional status with Nutritional Risk Score.</p> <p>Classification of pressure ulcers with NPUAP, EPUAP and PPPIA.</p> <p>SWC: continuous ulcer assessment, blood tests, use of wound dressings, mobilization, cleansing of necrotic tissues.</p> <p>HFUS: once a day, 5 days a week.</p> <p>ES: once a day, for 50', 5 days a week.</p> <p>Duration of monitoring: 4 and 6 weeks.</p>	<p>for normality of distribution with Shapiro-Wilk W-test.</p> <p>Patient characteristics in the 3 groups were evaluated for heterogeneity using the two-sided Fisher's exact test, the Kruskal-Wallis test and the Kruskal-Wallis post-hoc test.</p> <p>Mean WSA before and after treatment were compared using the Wilcoxon signed-rank test.</p> <p>The level of significance was set at $p < 0.05$.</p>	<p>(cm²):</p> <p>SWC + US group: 10,86→3,69</p> <p>SWC + ES group: 7,48→2,65</p> <p>SWC group: 9,31→5,33 ($p < 0.0001$).</p> <p>The percentage reduction in the surface area of Pus (4 weeks):</p> <p>SWC + US group: 70,93%</p> <p>SWC + ES group: 68,3%</p> <p>SWC group: 39%</p> <p>The percentage reduction in the surface area of PUs (6 weeks):</p> <p>SWC + US group: 77,48%</p> <p>SWC + ES group: 76,19%</p> <p>SWC group: 48,97%</p> <p>Healing in 6 weeks:</p> <p>SWC + US group: 13/28 (46,43%)</p> <p>SWC + ES group: 15/29 (51,72%)</p> <p>SWC group: 7/31 (22,6%)</p>	<p>duration of the research.</p> <p>Patients were not blinded, control groups with sham ES and sham US were not created.</p> <p>The healing ulcers were significant in all three groups, but pressure ulcers decreased significantly more and faster with the two considered methods.</p>
3. Polak et al., 2014, Poland.	Evaluation and comparison of HFUS efficacy in stage II and III pressure ulcers in combination with standard wound	<p>Prospective, randomized, controlled clinical study.</p> <p>Elderly patients admitted to 4 centers (n=42), with PrUs (n=44).</p>	<p>Prognosis for pressure ulcers with Norton scale and Braden scale, nutritional status with Nutritional Risk Score.</p>	<p>Statistical analysis: StatSoft Polska Sp. z o.o. version 8.0.</p> <p>Patient characteristics: Shapiro-Wilk W-test.</p>	<p>Total average reduction in the surface area of PrUs (cm²):</p> <p>SWC + HFUS group:</p> <p>15,38→6,16 ($p = 0,000069$)</p> <p>SWC group:</p>	<p>The study lacked patient blinding and the non-application of sham HFUS to the control group.</p> <p>Probably dif-</p>

care (SWC) versus standard wound care only.	<p>Stage II: 32 (72.73%) Stage III: 12 (27.27%)</p> <p>SWC group: (n=22) with PrUs (n=23), Stage II: 18 and Stage III: 5.</p> <p>SWC+HFUS group (n=20) with PrUs (n=21), Stage II: 14 and Stage III: 7.</p> <p>Study duration: 2,5 years.</p>	<p>Classification of pressure ulcers with NPUAP and EPUAP.</p> <p>SWC: regularly assessment of wounds, blood analysis, use of wound dressings, mobilization, cleansing of necrotic tissues.</p> <p>HFUS: once a day, 5 days in a week.</p> <p>The healing progress of ulcers receiving SWC and SWC+HFUS was monitored for 6 weeks or until wounds closed.</p>	<p>Distribution homogeneity of patient characteristics in both groups: Fisher test κ and Mann-Whitney U test.</p> <p>The level of significance was set at $p \leq 0.05$.</p>	<p>11,08–8,28 ($p=0,0062$)</p> <p>The percentage reduction in the surface area of Pus (6 weeks): SWC + HFUS group: 68.80% SWC group: 37.24% ($p=0,047$)</p> <p>Weekly average reduction in the surface area of PrUs (cm²): SWC + HFUS group: 2.63 SWC group: 1.52 ($p = 0,07$)</p> <p>Stage II SWC + HFUS: 3,09 Stage II SWC: 1,08 ($p=0,045$)</p> <p>Stage III SWC + HFUS: 1,70 Stage III SWC: 3,44 ($p=0,65$)</p> <p>Healing in 6 weeks: SWC + HFUS group: 8/21 (38,09%) ($p = 0.062$) SWC group: 3/23 (13,04%) ($p = 0.999$)</p> <p>Stage II SWC + HFUS 50%: 11/14 Stage II SWC 50%: 7/18 ($p=0,035$)</p> <p>Stage III SWC + HFUS 50%: 4/7 Stage III SWC</p>	<p>ferent SWC due to 4 research centers.</p> <p>Ulcers in the SWC + HFUS group decreased more in size.</p> <p>The weekly healing rate of ulcers in the SWC + HFUS group was better than in ulcers in the SWC group, but not statistically significant. Only for ulcers Stage II it was statistically significant.</p> <p>More stage II ulcers of the SWC + HFUS group were reduced to the surface by at least 50% compared to the same ulcers of the SWC group.</p>
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					50%: 3/5 (p=0,999)	
4. Maeshige et al., 2010, Japan.	Investigation and evalua- tion of the effect of ultrasound in combination with stand- ard wound care with a wound dressing in the treat- ment of pressure ulcers.	<p>Pilot random- ized controlled clinical trial.</p> <p>Elderly neuro- logical patients (n=5), with PrUs Stage III and IV (n=7).</p> <p>PrUs without local wound infection and extensive ne- crotic tissue. Patients with- out diabetes mellitus type 2 and/or periph- eral arterial disease.</p> <p>Treatment group (US): 3 patients with 4 ulcers (ulcers 1- 4).</p> <p>Control group (sham US): 2 patients with 3 ulcers (ulcers 5- 7).</p> <p>One ulcer was not random- ized, but was the first to re- ceive in the BABA se- quence, with a view to deter- mining if the pilot was feasi- ble.</p>	<p>Classification of pressure ulcers with NPUAP.</p> <p>Application of US for 10', 5 days per week for 2-4 weeks.</p> <p>All patients received standard wound care throughout the study.</p> <p>Period A: Each ulcer received standard wound care only for 2-4 weeks.</p> <p>Period B: standard wound care + US or sham US for 2-4 weeks in an ABABA se- quence.</p> <p>The ulcers were random- ly assigned (via a toss of a coin) to the US group or the control group. The latter group received sham US at period B.</p>	<p>At each dress- ing change, the wound size and exu- date weight were meas- ured.</p> <p>The DESIGN (P) classifica- tion scale was used to evaluate the total char- acteristics of each PrUs:</p> <ul style="list-style-type: none"> • Depth • Exudate • Size • Infection • Granulation • Necrosis • Pocket <p>Wound size: A pen-tablet system (Intuos 3, Wacom, Japan) was used to meas- ure the wound. The wound shape was digitized and the area measured using Scion Image soft- ware.</p> <p>Exudate weight: This was measured by subtracting the weight of the dressing before appli- cation from the weight immediately after removal.</p> <p>Any dressings</p>	<p>Treatment group (US): Wound size: In ulcers 1 and 2, a marked re- duction in wound size was observed after 3-4 weeks of US treatment. In ulcer 3, a reduction was observed as soon as US treatment be- gan. In ulcer 4, no clear reduc- tion was seen.</p> <p>Exudate weight: In ulcers 1 and 2, exudates weight in- creased during US therapy. In ulcer 3, there was no such increase. In ulcer 4, exudate weight could not be meas- ured as it leaked from the dressing.</p> <p>Control group (sham US): Wound size: These ulcers showed no marked reduc- tion in wound size. One ulcer (ulcer 6) was withdrawn from the study after it increased in size.</p> <p>Exudate weight: In ul- cers 5-7, exu- date weight was dependent</p>	<p>Small sample size.</p> <p>The efficacy of US in the treatment of PrUs is not cer- tain.</p> <p>It seemed that the delivered intensity of US 0,5W/cm² can penetrate per- meable dress- ings that keep the wound moist and had a positive effect on healing.</p> <p>The implemen- tation of the US in conjunction with the stand- ard wound care could promote the healing of pressure ulcers.</p>

				that were soaked with urine were excluded from the analysis.	on wound size, no increase in weight was recorded.	
5. Serena et al., 2009, H.Π.A.	Evaluation of the effectiveness of non-touch low frequency ultrasound therapy (MIST Therapy) in reducing bacterial colonization in stage III pressure ulcers and promoting healing.	<p>Prospective experimental study.</p> <p>Patients with musculoskeletal disorders (mean age: 60 years) in 3 trauma care centers (n = 11).</p> <p>Chronic stage III pressure ulcers with a wound volume of up to 160 cm³ and bacterial measurements > 10⁵ CFU/g of tissue (without infection) were studied.</p> <p>(Colony Forming Units per gram of tissue: CFU/g of tissue)</p> <p>Patients without clinical symptoms of acute ulcer infection, without head or neck injury, without malignancy in the wound bed.</p>	<p>Assessment of the bacterial load of the ulcer by quantitative tissue biopsy before and 2 weeks after MIST Therapy.</p> <p>MIST Therapy: 3 times per week for 2 weeks.</p> <p>Average duration of each treatment: 4'</p> <p>All patients underwent the standard wound care (SWC) during the study.</p> <p>(SWC: appropriate supportive surface, pressure relief, nutritional status control and dressings exudates management).</p> <p>Systemic and local antibiotics, local antiseptics, EMLA cream, antimicrobial dressings were not allowed.</p>	<p>Descriptive statistics were performed to summarize and compare the baseline data with results after 2 weeks of MIST Therapy.</p> <p>Bacterial quantities were summarized in CFU/g tissue.</p>	<p>The bacterial load was reduced from 4x10⁷ to 2x10⁷ after MIST Therapy.</p> <p>A different response of Gram- and Gram+ bacteria to ultrasound therapy was observed.</p> <p>The Staphylococcus species, including MRSA (Staphylococcus aureus resistant to methicillin) were significantly reduced after treatment. However, Streptococcus G showed only a moderate decrease and the number of Streptococci A increased in an individual.</p> <p>Healing in 2 weeks:</p> <p>The ulcer surface was reduced by 26% (13,8cm² → 10,8 cm²) and the ulcer volume was reduced by 20% (18,5cm³ → 11,6cm³).</p>	<p>Absence of the control group.</p> <p>The non-touch low frequency ultrasound (LFUS) treatment reduces the bacterial load in the ulcer bed promoting the healing.</p>