

Technomics Research, LLC.

Economic and Statistical Analysis of Healthcare Technology

1815 Medina Road Medina, MN 55356
PHONE: 763.473.6374 FAX: 763.473.8082
www.TechnomicsResearch.com

REPORT:

Effectiveness of the IYIA O₂Misly™ WTS-1000 Compared to Standard Wound Care

Prepared for

Adrian Pelkus
CEO

IYIA Technologies, Inc.
San Marcos
California

By

Technomics Research, LLC

Melissa Martinson, Ph.D., M.S.
Vice President
Technomics Research, LLC

04 October 2007

TABLE OF CONTENTS

1.0 EXECUTIVE SUMMARY2

2.0 INTRODUCTION.....3

3.0 DESCRIPTION OF THE O₂MISLY STUDY3

 3.1 PATIENT POPULATION.....3

 3.2 TREATMENT PROTOCOL3

 3.3 WOUND ASSESSMENT PROTOCOL4

4.0 DESCRIPTION OF THE STANDARD-OF-CARE STUDY4

5.0 STATISTICAL ANALYSIS METHODS4

 5.1 ANALYSIS OF O₂MISLY DATA4

 5.2 COMPARISON OF O₂MISLY TO STANDARD CARE META-ANALYSIS.....5

6.0 RESULTS OF THE ANALYSIS5

 6.1 RESULTS OF THE O₂MISLY ANALYSIS5

 6.2 RESULTS OF THE COMPARATIVE ANALYSIS9

7.0 CONCLUSIONS10

APPENDIX 1: O² MISLY PROTOCOL1

APPENDIX 2: MARGOLIS META-ANALYSIS.....1

1.0 EXECUTIVE SUMMARY

Nearly 2.5 million diabetic foot ulcers are diagnosed each year in the United States. These wounds are notoriously slow to heal, with many taking more than six months to resolve and many others resulting in amputation because of failure to heal.

IYIA Technologies has developed the O₂Misly™ Wound Treatment System (WTS) which delivers hyperoxia treatment followed by a humid mist, to which the physician can add his/her choice of antibiotic therapies. It is used as an adjunctive therapy to standard wound care.

A clinical study of the O₂Misly therapy included 31 patients with a total of 38 ulcers. All wounds had failed to heal with standard therapy, which consisted of debridement, avoidance of weight bearing, and moistened gauze. Twenty-three (23) of the patients had 29 diabetic neuropathic ulcers; 21 were males and 2 were females with an average age of 61. Of the 29 neuropathic ulcers, 19 were grade 2, six (6) were grade 3, and four (4) were reported as grade 3.5 by the University of Texas Health Science Center classification system. These wounds averaged 1181 mm³ in size and 24 months in duration. At 12 weeks, 38.4% of the wounds were healed. The proportion healed at 20 weeks was 46.0%.

For comparison, healing rates of uninfected, non-ischemic, neuropathic foot ulcers using standard wound care (as described above) were taken from a meta-analysis of randomized control trials. For the four control groups with a 12-week endpoint, the weighted mean healing rate was 24.2% (n=164). In the 6 control groups with a 20-week endpoint, the weighted mean healing rate was 30.9% (n=458).

The proportions of wounds closed by 12 and 20 weeks from the O₂Misly analysis were compared to those of the meta-analysis using a standard χ^2 -test for the equality of proportions. At 12 weeks, 38% of neuropathic wounds treated with O₂Misly were closed compared to 24% of standard-care wounds (p=0.004). At 20 weeks, 46% of O₂Misly wounds were healed compared to 31% of standard-care wounds (p=0.001).

O₂Misly therapy doubles the healing rates of chronic diabetic neuropathic wounds at 12 and 20 weeks. Because of the clinically large improvement due to O₂Misly, the difference in rates was highly statistically significant in spite of the relative small number of O₂Misly patients.

2.0 INTRODUCTION

Nearly 2.5 million diabetic foot ulcers are diagnosed each year in the United States. These wounds are notoriously slow to heal, with many taking more than six months to resolve and many others resulting in amputation because of failure to heal.

In response to the need for more effective wound healing, IYIA Technologies has developed the O₂Misly™ Wound Treatment System (WTS) device for wound care. O₂Misly delivers hyperoxia treatment followed by a humid mist, to which the physician can add his/her choice of antibiotic therapies. It is used as an adjunctive therapy to standard wound care.

O₂Misly was approved by 510K application to the FDA without clinical data. Because healthcare payers need clinical data before they will cover new technologies, IYIA Technologies has conducted a clinical study of the effectiveness of O₂Misly therapy on diabetic wounds. This report details the results of the study, and provides a comparison of the effectiveness of wound healing with O₂Misly to that achieved with standard wound care.

3.0 DESCRIPTION OF THE O₂MISLY STUDY

The study of the effectiveness of O₂Misly in treating diabetic and other difficult wounds was conducted under Protocol 2005-01. A copy of the protocol can be found in Appendix 1 of this report.

3.1 Patient Population

Patients targeted for the study were aged 18 or older of either sex with chronic diabetic, arterial, venous and decubitus foot ulcers. Wounds graded at study entry according to the University of Texas Health Science Center classification as Grade 0, 1 and 2 were included. All wounds had failed to heal with standard therapy.

3.2 Treatment Protocol

Patients received the O₂Misly™ treatments dispensed from the WTS-1000, in addition to standard ulcer care treatments and dressings. Treatments with O₂Misly were to follow this procedure:

1. The patient's wound is undressed, debrided and cleansed.
2. The patient places his/her foot into a clear plastic bag in a tank on the front of the system.
3. The therapist places the plastic panels around the patient's calf.
4. The therapist places the chosen antibiotic (if any) into the dispensing chamber.
5. The warm mist is applied to the patient's foot (inside the bag) for 10 minutes.

6. Oxygen is introduced into the area around the patient's foot for five minutes under a very slight pressure.
7. Repeated application of mist and oxygen follow the first for a total of four cycles (one hour) of treatment.
8. The patient removes his/her foot for evaluation, drying and dressing.
9. Treatments are to be applied four times a week for the duration of the O₂Misly therapy (usually until wound closure).

3.3 Wound Assessment Protocol

Wound size was measured by placing a ruler on the surface at the widest part of the long axis, and again at the widest part of the short axis. The depth was measured by placing a probe perpendicular to the surface into the deepest part of the wound.

4.0 DESCRIPTION OF THE STANDARD-OF-CARE STUDY

The data on wound healing using standard wound care alone was taken from a meta-analysis of randomized control trials.¹ A copy of the paper is provided in Appendix 2.

The meta-analysis included studies of uninfected diabetic neuropathic foot ulcers with adequate perfusion. The analysis estimated the weighted mean percentage of wounds healed by 12 weeks and 20 weeks. Ten controls from these studies were used to estimate the effectiveness of standard care, which included debridement, avoidance of weight bearing, and either saline-moistened gauze or placebo gel and gauze. Grouped-data logistic regression was used to assess the impact of patient age, wound size (area), and wound duration on the percentage healed.

5.0 STATISTICAL ANALYSIS METHODS

5.1 Analysis of O₂Misly Data

Descriptive statistics were calculated for the patients' demographic variables and their pre-treatment wound status.

The percentage of wounds healed at 12 and 20 weeks was estimated using the Kaplan-Meier Product-Limit method of survival analysis. This was repeated under the following scenarios:

¹ Margolis, David J. MD; Kantor, Jonathan MA; and Berlin, Jesse A. SCD. Healing of Diabetic Neuropathic Foot Ulcers Receiving Standard Treatment: A meta-analysis. Diabetes Care 22: 692-695, 1999.

1. Using all of types of wounds of the patients in the study with standard censoring of observations. Amputations were considered to be unhealed wounds at the end of follow-up.
2. Using only the diabetic neuropathic wounds with standard censoring of observations. Amputations were considered to be unhealed wounds at the end of follow-up.

Finally, a Cox's proportional hazards survival analysis with regression covariates was run on all of the wounds to test whether any of the following factors affected wound-healing time: type of wound, age of patient, gender, grade of wound, and volume of wound. It was not possible to include the duration of the wound in this analysis because this variable was collected from patient recall, and much of it was missing.

All analyses were performed using STATA 9.0 (StataCorp LP, College Station TX).

5.2 Comparison of O₂Misly to Standard Care Meta-Analysis

The proportions of wounds closed by 12 and 20 weeks from the O₂Misly analysis of diabetic neuropathic wounds (Healing Time Analysis #2) were compared to those of the meta-analysis using a standard χ^2 -test for the equality of proportions. This was used as an approximation to a test of the equality of survival curves, which was not possible to calculate because the survivor function from the meta-analysis was not provided in the paper. However, it should be conservative because proportions estimated with survival analysis techniques are generally more precise than a proportion calculated as $p=k/n$, so the test based on the approximation will have lower power and be less likely to detect a difference between O₂Misly and standard care.

The analysis includes the estimated differences between O₂Misly and standard care at 12 and 20 weeks, the p-values for the equality of the differences, and the 95% confidence intervals.

6.0 RESULTS OF THE ANALYSIS

6.1 Results of the O₂Misly Analysis

Demographics

The O₂Misly study included 31 patients with a total of 38 ulcers. Twenty-three (23) of the patients had 29 diabetic neuropathic ulcers; 21 were males and 2 were females. Of the eight (8) patients with non-diabetic ulcers, there were two (2) males and two (2) females with pressure wounds, one (1) male with a puncture wound, and two (2) males and one (1) female with diabetic wounds that were non-neuropathic. All but one of the nine (9) of the non-neuropathic

wounds were grade 2; the other was grade 4. Of the 29 neuropathic ulcers, 19 were grade 2, six (6) were grade 3, and four (4) were reported as grade 3.5.

Patients with non-neuropathic ulcers averaged 64 years of age (range: 34 – 80); their wounds averaged 318 mm³ in size (range: 24 – 650) and 20 months in duration (range: 1 – 48). Patients with diabetic ulcers averaged 61 years of age (range: 40 – 80); their wounds were substantially larger, and averaged 1181 mm³ (range: 18 – 9100) and 24 months duration (range: 1.5 – 96).

Table 6.1-1 summarizes the descriptive statistics about the patients and wounds.

Table 6.1-1: Patient and Wound Characteristics

Characteristic	Neuropathic		Non-neuropathic	
N of patients	23		8	
N of ulcers	29		9	
Gender	21 Males	2 Females	5 Males	3 Female
Patient age (y)	Mean: 61	Range: 40 -80	Mean: 64	Range: 34 - 80
Wound duration (m)	Mean*: 24	Range: 1.5 – 96	Mean**: 20	Range: 1 - 48
Wound size (mm ³)	Mean: 1181	Range: 18 - 9100	Mean: 318	Range: 24 - 650

* based on 15 patients reporting values

** based on 4 patients reporting values

The total number of treatments patients received averaged 13.5 (range: 2 – 65). On average, wounds were treated in two (2) sessions per week, with a minimum of 1 and a maximum of three (3).

Healing Time Analysis 1: All Wounds

Table 6.1-2 shows the survivor function for all wounds in the O₂Misly study; this function is displayed in Figure 6.1-1. Note that the healed proportion is 1.00 minus the “survivor function” proportion in the table. Thus, at 12 weeks (highlighted), .233 (23.3%) of the wounds were surviving, so 76.7% of the wounds were healed. The estimated proportion healed at 20 weeks was 90.7%. (The time of 15 weeks is highlighted because a week only appears in the table if a healing occurred at that time. This is why 20 weeks is not listed.)

In this analysis, patients who were released from the study (i.e., they had treatments terminated) were censored at the time of their release. Reasons

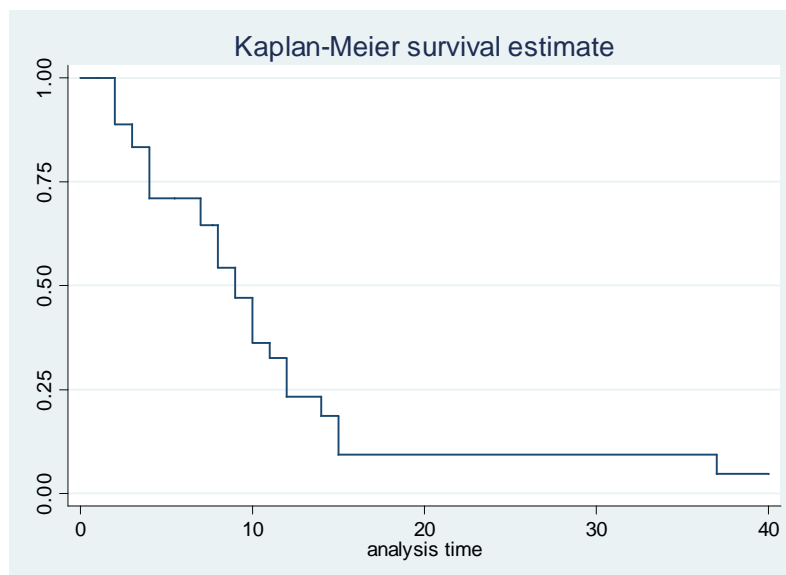
for release included the following:

1. Non-compliance: N=5; these patients refused to off-load the affected limb.
2. Surgery: N=2; one patient had bypass surgery and one had a foreign body removed from the wound (which turned out to be the reason that the wound would not heal).
3. Other: N=1; one patient had trouble arranging transportation to the treatment center.

Table 6.1-2: Healing Times for All Wounds

Time	Beg. Total	Fail	Net Lost	Survivor Function	Std. Error	[95% Conf. Int.]	
1	38	0	2	1.0000	.	.	.
2	36	4	0	0.8889	0.0524	0.7305	0.9568
3	32	2	3	0.8333	0.0621	0.6661	0.9214
4	27	4	0	0.7099	0.0778	0.5267	0.8326
5.5	23	0	1	0.7099	0.0778	0.5267	0.8326
7	22	2	0	0.6453	0.0830	0.4590	0.7817
7.7	20	0	1	0.6453	0.0830	0.4590	0.7817
8	19	3	1	0.5434	0.0883	0.3577	0.6965
9	15	2	0	0.4710	0.0902	0.2895	0.6329
10	13	3	0	0.3623	0.0886	0.1964	0.5308
11	10	1	2	0.3261	0.0868	0.1678	0.4948
12	7	2	0	0.2329	0.0833	0.0946	0.4064
14	5	1	0	0.1863	0.0786	0.0640	0.3580
15	4	2	0	0.0932	0.0609	0.0170	0.2509
37	2	1	0	0.0466	0.0449	0.0034	0.1908
40	1	0	1	0.0466	0.0449	0.0034	0.1908

Figure 6.1-1: Survival Time (w) of All Wounds



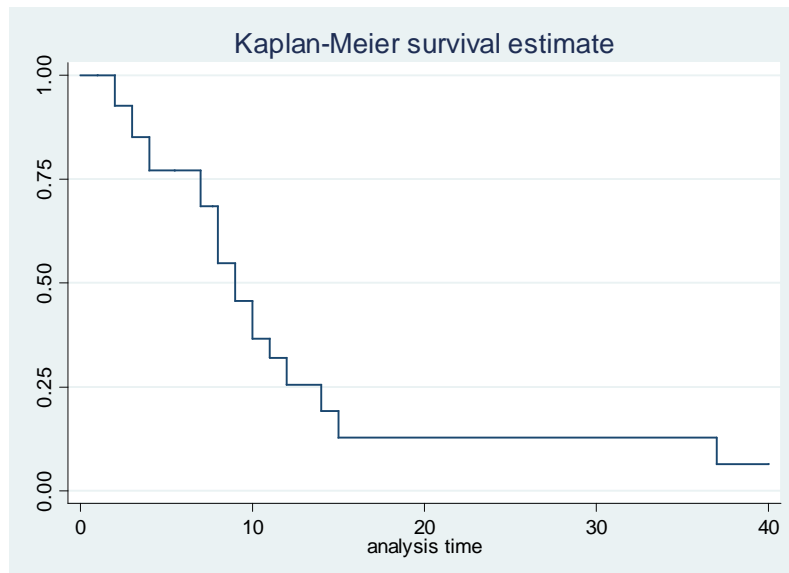
Healing Time Analysis 2: Diabetic Neuropathic Wounds

Table 6.1-3 shows the survivor function for diabetic neuropathic wounds in the O₂Misly study; this function is displayed in Figure 6.1-2. At 12 weeks (highlighted), .256 (25.6%) of the wounds were surviving, so 74.4% of the wounds were healed. The estimated proportion healed at 20 weeks was 87.2%. (The time of 15 weeks is highlighted because a week only appears in the table if a healing occurred at that time. This is why 20 weeks is not listed.)

Table 6.1-3: Healing Times for Diabetic Neuropathic Wounds

Time	Beg. Total	Fail	Net Lost	Survivor Function	Std. Error	[95% Conf. Int.]	
1	29	0	2	1.0000	.	.	.
2	27	2	0	0.9259	0.0504	0.7350	0.9809
3	25	2	2	0.8519	0.0684	0.6520	0.9417
4	21	2	0	0.7707	0.0825	0.5584	0.8901
5.5	19	0	1	0.7707	0.0825	0.5584	0.8901
7	18	2	0	0.6851	0.0929	0.4659	0.8292
7.7	16	0	1	0.6851	0.0929	0.4659	0.8292
8	15	3	0	0.5481	0.1026	0.3305	0.7213
9	12	2	0	0.4567	0.1039	0.2505	0.6417
10	10	2	0	0.3654	0.1012	0.1779	0.5559
11	8	1	2	0.3197	0.0983	0.1445	0.5106
12	5	1	0	0.2558	0.0973	0.0949	0.4541
14	4	1	0	0.1918	0.0916	0.0545	0.3919
15	3	1	0	0.1279	0.0803	0.0237	0.3230
37	2	1	0	0.0639	0.0605	0.0045	0.2463
40	1	0	1	0.0639	0.0605	0.0045	0.2463

Figure 6.1-2: Survival Time (w) of Diabetic Neuropathic Wounds



Analysis of Covariate Effects on Healing Time

The Cox's survival analysis with regression covariates did not demonstrate any significant relationships to healing time of all wounds: gender ($p=.656$); age ($p=.253$); grade ($p=.170$); wound type (diabetic neuropathic vs. other) ($p=.945$); and size ($p=.093$), although 'size' shows the possibility of affecting the healing time. The global χ^2 test for an improvement in fit due to all variables simultaneously was also not significant ($p=0.111$), indicating that a model with fewer of these variables would not show any to be significant. These negative results may be due to a lack of power.

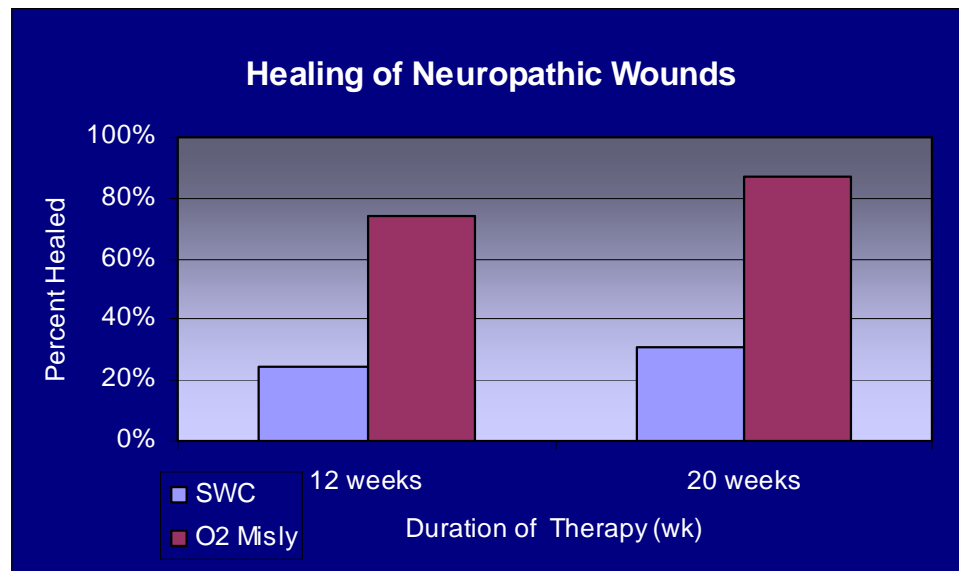
6.2 Results of the Comparative Analysis

The proportions of wounds surviving at 12 and 20 weeks were compared under all three analysis scenarios: all wounds, neuropathic wounds, and neuropathics wounds using worst-case healing results. In every analysis, O₂Misly therapy was superior to standard care in the proportion of wounds healed. Table 6.3-1 shows the estimates for each therapy, the difference with its 95% confidence interval, and the p-value for the comparison. In all cases, the p-value is highly statistically significant. Figure 6.2-1 shows the differences in the percentages of healed wounds at 12 and 20 weeks.

Table 6.2-1: Comparisons of O₂Misly and Standard Therapies

Statistic		Comparison	
		All Wounds	Neuropathic Wounds
12 Weeks	O ₂ Misly healing rate	76.7%	74.4%
	Standard healing rate	24.2%	24.2%
	Difference	52.5%	50.2%
	95% Confidence Interval	36% - 69%	33% - 67%
	p-value	< 0.0001	< 0.0001
20 Weeks	O ₂ Misly healing rate	90.7%	87.2%
	Standard healing rate	30.9%	30.9%
	Difference	59.8%	56.3%
	95% Confidence Interval	48% - 71%	42% - 69%
	p-value	< 0.0001	< 0.0001

Figure 6.2-1: Percent Healed Among of Diabetic Neuropathic Wounds



7.0 CONCLUSIONS

The comparison of the O₂Misly results to the standard-of-care results showed a significant improvement in neuropathic wound healing with O₂Misly. The proportions of healed wounds were about 200% better at 12 and 20 weeks: 24% compared to 74% and 31% compared to 87%. This is such a substantial improvement that even with a relatively small number of O₂Misly patients, the comparisons were highly statistically significant.

Of special interest is the fact that the O₂Misly wounds were healed after having failed standard wound care. This means that if O₂Misly were used the same way in real-world situations, one might expect 31 patients out of 100 to heal with standard care, and 87% of the remaining 69 patients, or 60 additional patients, to heal with O₂Misly. This strategy would heal a total of 91 of 100 patients.

Another piece of evidence supporting the effectiveness of O₂Misly therapy is anecdotal, but compelling. One patient on therapy showed substantial improvement over a several-week period. Then he was unable to receive therapy for some weeks, and his wound regressed beyond its original size. At that point, he committed to receiving regular O₂Misly therapy, and his wound healed within 20 weeks.

The major shortcoming of the comparison is that the patients were not randomized to their respective treatments. The standard-of-care patients were randomized against other wound-healing treatments, not against O₂Misly. Of course, the relevant question is whether the patients in the control group would have had the same results as the O₂Misly patients had they received O₂Misly therapy, or equivalently, whether the two groups contained comparable patients.

In both studies, the wounds were diabetic neuropathic wounds, and the standard-of-care described in the meta-analysis was essentially the same therapy that the O₂Misly patients received along with the new treatment. The control wounds were likely to have received the best available non-investigational care, as they were enrolled in randomized control trials. On other factors, it is not possible to assess the precise similarity, although it seems likely that the O₂Misly patients' wounds were at least as difficult to treat as the control patients':

- The control wounds were all uninfected; there was at least 1 and possibly 3 infected wounds in the O₂Misly study (1 grade 4 and 2 grade 3.5).
- The control wounds were all adequately perfused, so the control healing rate does not include patients who needed reperfusion therapies. In the O₂Misly study, several patients were sent for reperfusion therapy; these patients were censored in the O₂Misly analysis.
- The control wounds may not have all been chronic (wound duration was not given in the meta-analysis). In the O₂Misly study, all of the wounds were chronic and had previously failed the therapy that the control patients were receiving.
- The control wounds may not have been as large (size was not given in the meta-analysis). In the O₂Misly study, the average wound size was 1181 mm³; the largest was 9100 mm³. These are fairly large wounds.

From these considerations, it is clear that O₂Misly therapy represents an important improvement in diabetic wound care. Even challenging wounds can be effectively treated in a short period of time, avoiding long treatment periods, infections, and surgeries. It is likely that O₂Misly therapy will save payers money in addition to improving patient outcomes.

APPENDIX 1: O² MISLY PROTOCOL



IYIA Technologies, Inc. designs, manufactures, and markets high tech medical healing products.

PROTOCOL 2005-01

Trial of the Misly™ Wound Treatment System (WTS-1000) " When Added to the Standard of Care in the Treatment of Non-healing Foot Ulcers.

Misly™ is an adjunctive therapy, not the sole method of treatment. The Misly™ WTS-1000 enhances wound Treatment by hydrating and helping increase tissue oxygenation. Adequate tissue oxygenation and hydration promotes revascularization and collagen deposition. To aid in fighting infections, anti-bacterial ingredients can be added.

Summary

Patients who will receive the Misly™ Treatments dispensed from the WTS-1000, in addition to standard ulcer care treatments and dressings, will be asked to place their foot into a clear plastic bag that is in a tank on the front of the system. Plastic panels will be placed around the patient's calf. These have a comfortable foam liner that will encircle the patient's calf. A warm mist will enter the bag surrounding the patient's foot. The hydrating mist contains an anti-bacterial. After 10 minutes the mist will stop and Oxygen will be inserted into the area around the patient's foot for 5 minutes under a very slight pressure. Repeated application of mist and oxygen will follow the first for a total of four cycles, total 1 hour per treatment. The plastic panels will be removed and the patient will be asked to remove their foot for evaluation, drying and dressing. The process will be applied four times a week for the duration of the treatment, not to exceed ten weeks.

Intended Use of the Wound Treatment System WTS-1000

The intended use of Misly™ is to provide humidified hyperbaric oxygen to open, chronic wounds as an adjunct therapy in wound management and treatment. Increasing the oxygen concentration above chronic

open wounds with the use of this topical hyperbaric oxygen device may promote the rate of Treatment and suppress bacterial growth.

The Device is intended for the following kinds of open acute or chronic wounds:

- Skin ulcerations due to diabetes, venous stasis, post surgical infections and gangrenous lesions,
- Decubitus ulcers
- Amputations / infected stumps
- Skin grafts
- Burns
- Frostbite

Contraindications

- Acute skin conditions
- Inadequate perfusion to support Treatment
- Wounds where the end cannot be probed
- Wounds covered with petroleum based dressing
- Non-compliant patient

This protocol is intended for professional clinicians to ensure the proper use of the Misly™ WTS-1000.

1. Screening Visit (as explained to the patient)

If you decide to participate in the study, the following events will occur:

- Your medical history will be obtained, including information about your previous foot ulcer care and the circumference of you calf measured 5" up from the ankle.
- You will have a physical examination including blood pressure, temperature, and pulse. Your wound severity will be classified.
- You will have a painless test performed to see how much feeling you have in your feet. This test (called a monofilament test) is done by "whispering" the foot with a special instrument in order to evaluate sensory impairment.
- You will be asked about any medications you are taking.
- If you have a dressing on your ulcer, it will be removed and your ulcer will be examined, cleaned, and measured.

- The unhealthy tissue from your ulcer will be surgically removed from the ulcer. Pictures will be taken of the ulcer before and after the unhealthy tissue is removed. Your study doctor may remove unhealthy tissue or you may be referred to another doctor for this procedure. You will be advised of these plans and where the procedure will be performed.
- After the unhealthy tissue is removed from your ulcer, a tissue biopsy for culture (a test to check for infection) will be obtained from your ulcer to determine if infection is present.
- An x-ray of the ulcer area may be performed.
- A PO2 blood test may be performed.
- Ankle brachial index and toe brachial index will be performed. These painless tests include checking your arm blood pressure and then repeating the test just above your ankle and at your toe. The doctor interprets the results between the two test sites. A sound wave probe will also be used to assess blood flow to your feet.
- You will receive instructions on the care of your ulcer and dressings and relieving pressure on your foot between visits to the study doctor. You will also be encouraged to eat your meals and take all of your medicines correctly.
- You will be asked about your care at home, your blood sugar levels, and any problems with diabetes care. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.

2. Seven (7)-Day Screening Period (as explained to the patient)

After the screening procedures and the surgical removal of tissue to test for infection (also called debridement) have taken place and the study doctor agrees that you are eligible to continue with the screening process, the following events will occur:

- Your ulcer will be dressed (bandaged).
- You will be given special footwear, offload device, or a wheel chair to help keep pressure of the ulcer area.

You will be asked to return to the study doctor in 6-8 days so the doctor can determine if you are eligible to enroll into the study. You will return 1 or 2 times within 8 days for dressing change.

3. Randomization Visit (as explained to the patient)

After the screening procedures are completed, including the 7 day screening period, and the study doctor agrees that you may be enrolled in the study, the following events will occur:

- You will be assigned (like flipping a coin) to be in either the Misly Wound Treatment group or the standard ulcer care group and you will have your ulcer cared for according to your study group assignment.
- Your ulcer dressing will be removed and your ulcer will be examined, cleaned, and measured.
- If the study doctor thinks that it is necessary, unhealthy ulcer tissue will be surgically removed from the ulcer.
- Pictures of the ulcer will be taken. These pictures will be taken when your ulcer is evaluated.
- If the study doctor thinks it is necessary, a swab (culture) will be taken of your ulcer to check for infection.
- The Misly Treatment or standard treatment for your group will be applied.
- A PO2 blood test may be performed.
- You will be asked about any problems or bad effects you have experienced since your last visit.
- You will be asked about your care at home, your blood sugar levels, and any problems with diabetes care since your last visit. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.
- You will be asked about your use of the footwear, crutches, walker or wheelchair since your previous visit.
- You will receive instructions on the care of your ulcer and dressings and relieving pressure on your foot between visits to the study doctor. You will also be encouraged to eat your meals and take all of your medicines correctly.
- You will be asked about any new medicines you are taking or any changes in the medications you were taking when you last visited the study doctor.
- If you are a female and able to become pregnant, a urine pregnancy test may be performed if you believe you are pregnant or you have symptoms suggesting a pregnancy.

4. Treatment Visits (as explained to the patient)

Regardless of which group you are assigned to, your ulcer will be evaluated by the study staff two times a week for twelve weeks or until your ulcer is healed, whichever comes first. During these evaluations, the following events will occur:

- Your weight, blood pressure, heart rate, and temperature will be measured.
- Your dressing will be removed and your ulcer will be examined, cleaned, and measured.
- If necessary, unhealthy ulcer tissue will be surgically removed from the ulcer.
- Pictures of the ulcer will be taken. These pictures will be taken when your ulcer is evaluated and if unhealthy tissue is removed, the pictures will be taken after this procedure is completed.
- The study device or standard bandage for your group will be applied and the dressing replaced.
- You will be asked about any excessive bleeding, problems, or bad effects you have experienced since your last visit.
- You will be asked about your care at home, your blood sugar levels, and any problems with diabetes care since your last visit. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.
- You will be asked about your use of the footwear or wheelchair since your previous visit.
- You will receive instructions on the care of your ulcer and dressings (bandages) and relieving pressure on your foot between visits to the study doctor. You will also be encouraged to eat your meals and take all of your medicines correctly.
- If necessary, an x-ray and a swab for culture may be taken of your ulcer.
- Your PO₂ level may be measured.
- If you are a female, and able to become pregnant, urine pregnancy test may be performed if you believe you are pregnant or you have symptoms suggesting a pregnancy.
- You will be asked of any new medications you are taking or any changes in the medications you were taking when you last visited the study doctor.

Proceeding with the Misly™ treatment:

1. Gather supplies.
2. Wash hands, don gloves.

3. The Universal No Smoking Sign on the side of the WTS-1000 must be clearly visible.

a) Remove all highly combustible material from the room.

b) Place any electrical appliance (e.g. television) at least five feet from the patient.

c) Move equipment or personal items (e.g. clothing, jewelry) likely to produce static discharges at least five feet from the patient.

4. Remove old dressing and expose wound. Cleanse first then debride. The wound base must be clean and dry. Photograph wound.

5. Gently guide the patients foot into the bag suspended in the WTS-1000 treatment tank. Take similar care when removing foot from the WTS-1000.

6. Attach the lid pieces with the disposable foam cuffs in place around the calf. Close lid around calf with the foam cuffs fit for comfort, no openings should be visible.

7. Open the valve to allow the hydrating mist to enter the bag. The vapor will be visible through the lid. Watch the temperature and humidity gauge mounted under the lid. Usual temperature is 80-85°f. Humidity 90%+ Stop treatment if patients feels discomfort. Allow vapor to fill bag for 10 minutes then close valve.

8. For Topical Hyperbaric Oxygen Treatment, connect the WTS-1000 to the oxygen inlet tube (green end) from the oxygen regulator and open valve to start flow at 10 liters per minute.

Fill the bag until taut. Fitted foam cuffs act as a pressure relief valve preventing over inflation. Monitor pressure on gauge mounted to lid. Assure pressure stays below 22mmHg / .4 psi.

Oxygen flow can be stopped, or continued at low level to ensure proper inflation is maintained. Administer oxygen for 5 minutes checking for leaks and resealing if necessary. Add additional oxygen if necessary to maintain inflation and keep bag taut.



WARNING: GREATER THAN 22 mm Hg OF MERCURY OXYGEN PRESSURE IN BAG MAY OCCLUDE ARTERIAL CIRCULATION, WHICH MAY LEAD TO A DECREASE IN LOCAL TISSUE CIRCULATION.

9. Repeat hydration / oxygenation cycle four times. Remove foot, dry, inspect, photograph and dress wound.

5. End of Treatment (as explained to the patient)

Subjects with a Study foot wound that is closed within the 12-week treatment phase will return one-week later to confirm the wound closure. The following events will occur:

- Your weight, blood pressure, heart rate, and temperature will be measured.
- Your ulcer will be examined and measured.
- If you are female and able to become pregnant, a urine pregnancy test may be performed if you believe you are pregnant or you have symptoms suggesting a pregnancy.
- You will have a picture taken in the area of your ulcer.
- Your ulcer will be dressed (bandaged) as needed. The standard wound treatment for the study will be applied.
- You will be asked of any bad infections you have had since your last visit.
- You will be asked about your home care, your blood sugar levels, and any problems with diabetes care since your last visit. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.
- You will be asked if you have been using the footwear, crutches, walker or wheelchair correctly.
- A member of the study staff will review your medical records.
- A culture of your ulcer will be taken, if necessary.
- You will be asked about any new medications you are taking or any changes in the medications you were taking when you last visited the study doctor.
- You will receive instructions on the care of your ulcer and dressings and relieving pressure on your foot between visits to the study doctor. You will also be encouraged to eat your meals and take your medicines correctly.
- If the study foot wound is not closed, the subject will continue care as randomized through normal course of treatment or presumed closure. This process will be repeated as necessary through week 12 or confirmation of closure.

6. Post-Closure Follow-Up Visits (as explained to the patient)

If your ulcer closes during the treatment period (between 1 and 12 weeks after your first treatment) and the ulcer has remained closed at the time of the End of Treatment Visit, you will return for three follow-up visits so the study doctor can evaluate whether your ulcer is still closed. These visits will

take place over an 11 week period. The following events will occur at each visit:

- Your weight, blood pressure, heart rate, and temperature will be measure.
- You will have a picture taken of your ulcer.
- Your ulcer area will be examined and measured.
- Your PO2 may be measured.
- Your ulcer will be dressed (bandaged) as needed.
- You will be asked about any excessive bleeding or bad effects you have had since your last visit.
- You will be asked if you have been using the footwear or wheelchair correctly.
- You will be asked about your care at home, your blood sugar levels, and any problems with diabetes care since your last visit. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.
- You will be asked about any new medications you are taking or any changes in the medications you were taking when you last visited the study doctor.
- You will receive instructions on the care of your ulcer and dressings and relieving pressure on your foot between visits to the study doctor. You will also be encouraged to eat your meals and take all of your medicines correctly.

If your ulcer has re-opened, you will be withdrawn from the study and given instructions for future care.

7. End of Study Visit/Procedures (as explained to the patient)

You will have an End of Study Visit in the case of any of the following circumstances:

- If your ulcer has not healed within 12 weeks after your first application of study device, end of study procedures will be performed at your week 12 visit.
- If your ulcer heals within 12 weeks after your first application of study device and you have entered the follow-up phase of the study, end of study procedures will be performed at the final follow-up visit.

- If your ulcer reopens during the follow-up phase, end of study procedures will be performed at that time.
- If you withdraw from the study or are terminated from the study by the study doctor, end of study procedures will be performed.

The following events will occur at the End of Study Visit:

- Your weight, blood pressure, pulse, and temperature will be measured.
- You will have a physical exam including blood flow assessment to your feet.
- You will have a painless test performed to see how much feeling you have in your feet. This test (called a monofilament test) is done by “whispering” the patient with a special instrument in order to evaluate sensory impairment.
- You will have a picture taken of the area of your ulcer.
- Your ulcer area will be examined and measured.
- Your ulcer will be dressed (bandaged) as needed..
- If you are a female and able to become pregnant, a urine pregnancy test may be performed if you believe you are pregnant or you have symptoms suggesting a pregnancy.
- You will be asked about any problems or bad effects you have experienced since your last visit.
- You will be asked your use of the footwear, crutches, walker or wheelchair since your previous visit.
- You will be asked about your care at home, your blood sugar levels, and any problems with diabetes care since your last visit. If you do not conduct your own blood glucose monitoring at home, the study doctor will perform a random finger stick blood glucose test during your visit.
- You will be asked about any new medications you are taking or any changes in the medications you were taking when you last visited the study doctor.

APPENDIX 2: MARGOLIS META-ANALYSIS