

Improvement of Cutaneous Sensitivity in Diabetic Peripheral Neuropathy With Combination L-Methylfolate, Methylcobalamin, and Pyridoxal 5'-Phosphate

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Studies of monotherapy with L-methylfolate, methylcobalamin, or pyridoxal 5'-phosphate suggest that these B vitamins may reverse both the symptoms and the pathophysiology of diabetic peripheral neuropathy (DPN). The efficacy of oral-combination L-methylfolate, 3 mg; methylcobalamin, 2 mg; and pyridoxal 5'-phosphate, 35 mg (LMF-MC-PP) in restoring cutaneous sensitivity in patients with type 2 diabetes with DPN was evaluated in 20 type 2 diabetic patients who were given LMF-MC-PP twice daily for 4 weeks and then once daily for an additional 48 weeks. Statistically significant improvement in 1-point (tactile) and 2-point (discriminatory) static testing at the right and left great toe and heel in the patients was observed in all 3 follow-up periods: 1) baseline to 6 months, 2) baseline to 1 year, and 3) 6 months to 1 year. The greatest improvement occurred between baseline and 1 year of treatment. Treatment with oral LMF-MC-PP appears to promote restoration of lost cutaneous sensation in DPN.

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Diabetic peripheral neuropathy (DPN) affects up to 60% of patients with diabetes mellitus (DM) and is a leading cause of disability in the United States. Approximately 30% of DM patients at least 40 years of age have DPN-related loss of protective sensation in their feet. This symptom precedes most foot ulcerations and nontraumatic amputations in DM patients.¹ In 2004,

more than 70,000 nontraumatic lower-limb amputations (representing approximately 60% of all nontraumatic amputations) were performed in patients with DM.²

The gravity of DPN is underscored by the fact that DM has reached epidemic proportions within the past quarter century in the United States. Between 1992 and 2002, the number of individuals diagnosed with DM doubled. In 2007, the estimated prevalence of DM in the general US population was 7.8%—a total of 23.6 million people. Of this group, DM remained undiagnosed in 5.7%.² The severity of DPN increases with duration of DM, patient age, and degree of glycemic dysregulation.^{3,4}

Despite the rising prevalence of DPN and the seriousness of its consequences, little consensus regarding its underlying pathophysiology and primary treatment exists.^{5,6} Although several conventional anti-seizure, antidepressant, and miscellaneous agents are used in treatment of DPN, only duloxetine and pregabalin have been approved by the US Food and Drug Administration (FDA) for the treatment of DPN pain and

pathologic processes associated with the disorder have prompted research into new pharmacologic interventions. For example, oxidative stress and nerve perfusion deficits have emerged as leading candidates in the pathogenesis of DPN. A recently proposed model suggests that hyperglycemia-induced oxidative stress and secondary depletion of potent endogenous vasodilating nitric oxide (NO) occur within the endoneurial microvasculature and, thus, the peripheral nerve trunks of patients with DPN. This pathology has been postulated to underlie the diabetic foot.⁷

L-Methylfolate, methylcobalamin, and pyridoxal 5'-phosphate are

L-methylfolate, 2 mg of methylcobalamin, and 35 mg of pyridoxal 5'-phosphate (LMF-MC-PP; Metanx®; PamLab, LLC, Covington, LA)¹³ improves sensory perception, as measured by 1-point (tactile) and 2-point (sensory discrimination) tests, in symptomatic type 2 DM patients with established peripheral neuropathy.

Patients and Methods

Patients

Twenty consecutive patients with type 2 DM with complaints consistent with DPN were screened at the Carolina Medical Associates Podiatric Clinic (Aiken, SC) between June 2006 and October 2007. Written

L-methylfolate, methylcobalamin, and pyridoxal 5'-phosphate are metabolically active forms of the B vitamins folate, B₁₂, and B₆, respectively.

metabolically active forms of the B vitamins folate, B₁₂, and B₆, respectively. Research has demonstrated that 5-methyltetrahydrofolate (5-MTHF) improves bioavailability of NO and, presumably, endothelial function in patients with type 2 diabetes.⁸

informed consent was obtained from all patients.

Inclusion Criteria

1) Positive sensory symptoms (eg, spontaneous pain or painful dysesthesias) of the feet for > 6 months (all study participants had spontaneous pain in the feet that was described as burning, tingling, throbbing, or like "pins and needles," consistent with diabetic neuropathy. At time of presentation, each patient had been evaluated for degree of neuropathy by means of monofilament [Semmes-Weinstein] testing. Each patient had failed at least 2 [ie, 20%] of 10 sites tested on each foot. At baseline in the study, each patient underwent a detailed neurologic examination, including assessment of skin condition, deep tendon reflexes, and muscle strength, as well as testing of touch, pinprick, proprioceptive, and vibratory sensation, and symptoms of neuropathy with a visual analog scale [VAS]); and

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dysesthesias. Unfortunately, despite the ability of these 2 drugs to reduce these symptoms, neither alters the underlying pathophysiology of DPN. To date, available conventional agents used in DPN are merely palliative, rather than disease modifying. Therefore, agents that address the underlying pathophysiology of DPN are needed.⁷

Although decades of intense research have produced no clear understanding of the pathophysiology of DPN, commonalities among several

Additional benefits of L-methylfolate may include a direct effect on endothelial NO synthase and a scavenging effect on free radicals, including superoxide anion and peroxynitrite.⁹ Oral methylcobalamin has been shown to enhance myelin node genesis, nerve regeneration, and improvement of paresthesias and dysesthesias in DPN.^{10,11} Also, there is a correlation between pyridoxal deficiency and symptomatic DPN.¹²

We hypothesized that twice-daily oral administration of 3 mg of

2) Willingness to participate in a 1-year study to evaluate response to medication; and

3) Active engagement with a primary-care physician for type 2 DM management. Although it was required that glycemic control was being attempted, patients were not required to have a specific average fasting blood glucose or hemoglobin A_{1C} to be enrolled in the study.

Exclusion Criteria

1) Failure to keep scheduled appointments for Pressure-Specified Sensory Device (PSSD; Pressure-Specified Sensory Device™; NK Biotechnical Corp., Minneapolis, MN) testing in any scheduled 2-week assessment period; and/or

2) Missing 3 consecutive doses of drug; and/or

3) Initiation of on-label or off-label medications (eg, gabapentin, pregabalin, or duloxetine) for diabetic neuropathy during the treatment period. (Discontinuation of any of these medications during the treatment period did not result in exclusion of a patient from study. Use of pregabalin, duloxetine, or amitriptyline at onset of study did not result in exclusion of a patient from the study, as these agents do not affect sensory capacity); and/or

4) Comorbidity for neuropathy or history of back surgery.

Methods

At baseline, each patient was evaluated at right and left great toe pulp and right and left medial heel (at the medial tuberosity) with the PSSD. The great toe and medial heel are innervated by fibers from the medial plantar nerve and the calcaneal branch of the lateral plantar nerve, respectively.

Following baseline PSSD testing, patients were treated with LMF-MC-PP twice daily for 4 weeks and then once daily for an additional 48 weeks. Samples of the medication were dispensed in sufficient quantity at each patient visit to allow for daily dosing beyond the next scheduled appointment. Patients were evaluated every 6 to 8 weeks for changes in symptoms, use of other medications, and adherence to the study protocol.

After 6 months and 1 year of treatment, patients underwent follow-up PSSD testing. In the current study, the following 4 test outcomes were measured. The mean differences in results were expressed in g/mm², for 1) combined (right and left) great toe pulp 1-point static PSSD testing, 2) combined heel 1-point static testing, 3) combined great toe 2-point static testing, and 4) combined heel 2-point static testing over 3 predetermined time intervals (baseline to 6 months, baseline to 1 year, and 6 months to 1 year). One-point static touch measures overall pressure threshold; 2-point static touch measures nerve density, which correlates with the number of surviving A-beta fibers in the dermis.

Statistical Analysis

The research question that was posed was whether the combined (average of right plus left) measures for great toe or heel for 1-point or 2-point static test result was significantly different either at 6 months of treatment as compared with baseline, at 1 year of treatment as compared with baseline, or at 1 year of treatment as compared with 6 months of treatment with oral LMF-MC-PP. Statistical analysis was conducted by Baylor Health Systems (Dallas, TX) using SAS® 9.2 software (SAS Software, Cary, NC). The 1-sample *t*-test, which was based on the difference in combined (right and left) static

values across any 2 time points, was used for data analysis. The rate of Type 1 error (mistakenly rejecting true null hypothesis) of each *t*-test was set at 0.0166. Thus, the overall Type 1 error rate for each static or combined static was controlled below 0.05. This maneuver is known as the Bonferroni adjustment for multiple testing. Results of the 1- or 2-point static test were expressed in mean change over the 3 time intervals, *P* value, and 98.34% confidence interval (CI) of each comparison. For patients with total loss of protective sensation at baseline, 130 g/mm² was used as the imputing value for their PSSD baseline measurement.

Results

PSSD Test Results

Mean results of PSSD test at baseline. Table 1 lists the mean absolute PSSD test results on the great toe pulp and medial heel of the patients at baseline, 6 months of treatment, and 1 year of treatment with LMF-MC-PP. At baseline, the following mean results were obtained:

Combined 1-point test on great toe pulp: 38.5 g/mm²

Combined 2-point test on great toe pulp: 49.5 g/mm²

Combined 1-point test on medial heel: 33.6 g/mm²

Combined 2-point test on medial heel: 53.3 g/mm²

Figures 1 and 2 illustrate the decline in absolute PSSD 2-point static test results (showing improvement in 2-point sensory discrimination) at the great toe pulp and medial heel, respectively, at baseline, 6 months, and 1 year. Overall, the greatest degree of improvement occurred between baseline and 1 year of treatment. Least improvement in PSSD results was observed between 6 months and 1 year of treatment.

Table 1
Absolute PSSD Test Results for Combined 1- and 2-point
Great Toe Pulp and Medial Heel

PSSD Testing Site	Mean (g/mm ²)
Great Toe Pulp 1-Point Combined	
Baseline	38.5
6 mo	15.9
1 y	4.9
Great Toe Pulp 2-Point Combined	
Baseline	49.5
6 mo	27.7
1 y	10.8
Medial Heel 1-Point Combined	
Baseline	33.6
6 mo	14.8
1 y	5.5
Medial Heel 2-Point Combined	
Baseline	53.3
6 mo	23.0
1 y	11.4

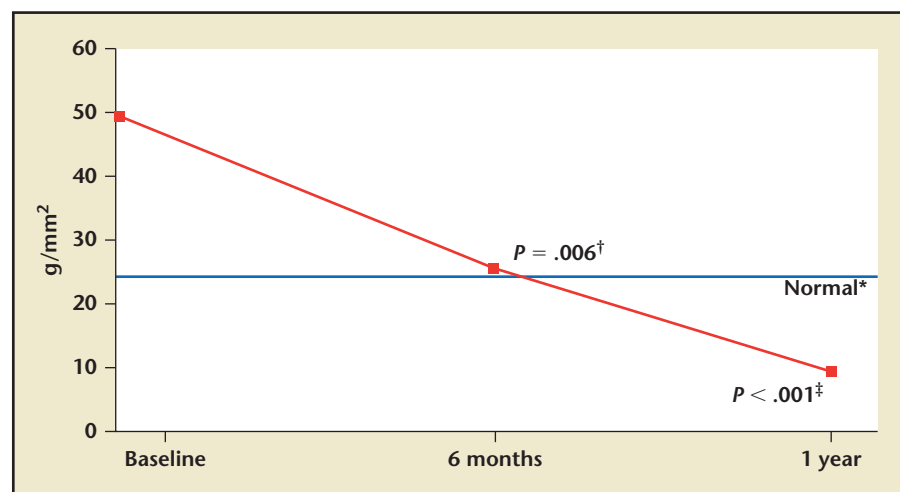
PSSD, Pressure-Specified Sensory Device™ (NK Biotechnical Corp., Minneapolis, MN).
 Data Courtesy of Mackie J. Walker, Jr, DPM (Carolina Musculoskeletal Institute, Aiken, SC).

Figure 1. Effect of oral LMF-PP-MC treatment on 2-point static testing on great toes (left/right combined) of patients with DPN. Results from testing the great toes of DPN patients ($n = 16$) with PSSD reveal decline in absolute PSSD 2-point static test values (showing improvement in 2-point sensory discrimination) at baseline, 6 months, and 1 year with oral-combination LMF-PP-MC. DPN, diabetic peripheral neuropathy; LMF-MC-PP, L-methylfolate, 3 mg; methylcobalamin, 2 mg; pyridoxal 5'-phosphate, 35 mg; PSSD, Pressure-Specified Sensory Device™ (NK Biotechnical Corp., Minneapolis, MN).

* ≤ 25.7 g/mm² represents the 99% confidence level for the normal pressure threshold, as measured by PSSD testing.

† Baseline vs 6 months of treatment with LMF-MC-PP.

‡ Baseline vs 1 year of treatment with LMF-MC-PP.



Mean changes in PSSD measurements.

Table 2 lists the mean changes in static 1- and 2-point PSSD tests of combined (right and left) great toe and medial heel that occurred during the 3 treatment periods: baseline to 6 months, baseline to 1 year, and 6 months to 1 year. One- and 2-point static PSSD values after 6 months and 1 year of treatment showed statistically significant improvement as compared with baseline values. Compared with baseline, the average changes in 2-point static sensory discrimination at the great toe after 6 months and 1 year of treatment with LMF-MC-PP were -27.05 g/mm² ($P = .006$) and -38.56 g/mm² ($P < .001$), respectively. Compared with the baseline, the average changes in 2-point static sensory discrimination at the medial heel after 6 months and 1 year of treatment with LMF-MC-PP were -34.1 g/mm² ($P < .001$) and -42.72 g/mm² ($P < .001$), respectively.

The majority of the patients who completed the VAS reported improved VAS scores in advance of improvement in PSSD scores. However, patients with loss of protective sensation and very low PSSD test results often reported an increase in unpleasant paresthesias for 3 to 8 weeks before improvement in PSSD results.

Discussion

Our study found that treatment with LMF-MC-PP was associated with statistically significant improvement in both 1- and 2-point static PSSD measurements in the great toe and heel at 6 months and 1 year. To our knowledge, this is the first clinical study to show that treatment with LMF-MC-PP was associated with statistically significant improvement in impaired peripheral nerve sensitivity due to DPN. These findings suggest that LMF-MC-PP promotes restoration

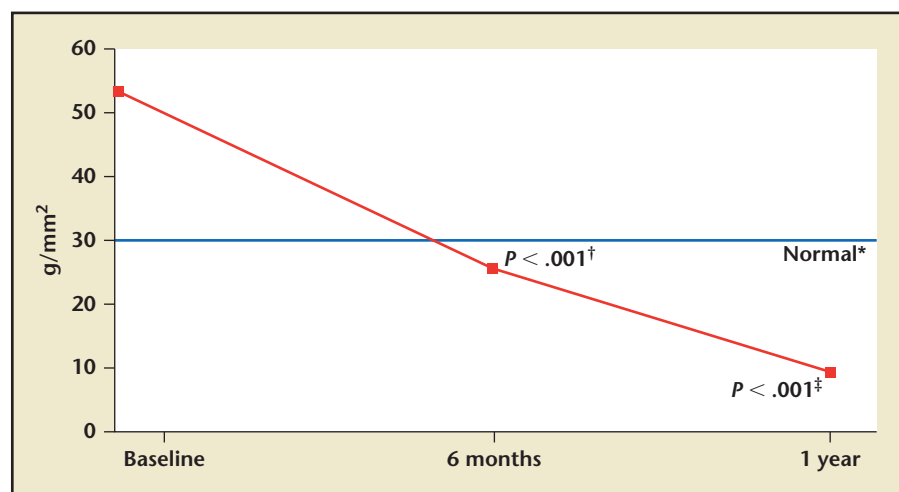


Figure 2. Effect of oral LMF-MC-PP treatment on 2-point static testing on medial heels (left/right combined) of patients with DPN. Results from testing the medial heels of DPN patients ($n = 16$) with the PSSD reveal decline in absolute PSSD 2-point static test values (showing improvement in 2-point sensory discrimination) at baseline, 6 months, and 1 year with oral-combination LMF-MC-PP. DPN, diabetic peripheral neuropathy; LMF-MC-PP, L-methylfolate, 3 mg; methylcobalamin, 2 mg; pyridoxal 5'-phosphate, 35 mg; PSSD, Pressure-Specified Sensory Device™ (NK Biotechnical Corp., Minneapolis, MN).

* ≤ 25.7 g/mm² represents the 99% confidence level for the normal pressure threshold, as measured by PSSD testing.

[†]Baseline vs 6 months of treatment with LMF-MC-PP.

[‡]Baseline vs 1 year of treatment with LMF-MC-PP.

of damaged cutaneous nerve fibers in patients with DPN.

The finding that patients with loss of protective sensation tended to have an increase in paresthesias for 3 to 8 weeks before improvement in PSSD test results suggests that a transient increase in paresthesias during treatment with LMF-MC-PP may be an early indicator of response to treatment. Patients reporting improvement in function tended to notice improvement in sleep first and then improvement in gait stability as paresthesias or pain declined, most often after 9 to 12 weeks of therapy.

Our findings are consistent with the theory that treatment with medical foods that enhance the bioavailability of endothelial NO may address the underlying pathogenesis of DPN. The

Table 2
Mean Differences in PSSD Measurements on Feet of Patients With Diabetic Peripheral Neuropathy

PSSD Test Site and Timing	Mean g/mm ²	T-Static	P Value	98.34% Confidence Intervals
Great Toe 1-Point Static				
Baseline vs 6 mo	-22.54	-2.93	.010 ^a	-43.25, -1.83
Baseline vs 1 y	-33.64	-3.99	.001 ^a	-56.35, -10.92
6 mo vs 1 y	-11.10	-2.57	.021	-22.73, 0.54
Medial Heel 1-Point Static				
Baseline vs 6 mo	-18.76	-3.09	.007 ^a	-35.10, -2.41
Baseline vs 1 y	-28.12	-4.38	< .001 ^a	-57.89, -19.23
6 mo vs 1 y	-9.36	-2.50	.025	-19.46, 0.74
Great Toe 2-Point-Static				
Baseline vs 6 mo	-27.05	-3.42	.006 ^a	-49.40, -4.69
Baseline vs 1 y	-38.56	-5.63	< .001 ^a	-57.89, -19.23
6 mo vs 1 y	-16.87	-3.49	.003 ^a	-29.87, -3.86
Medial Heel 2-Point Static				
Baseline vs 6 mo	-34.01	-5.93	< .001 ^a	-49.60, -18.41
Baseline vs 1 y	-42.72	-7.95	< .001 ^a	-57.34, -28.10
6 mo vs 1 y	-11.68	-2.33	.035	-25.23, 1.86

N = 16.

^aStatistically significant.

formulation of LMF-MC-PP used in the current study is classified by the FDA as a prescription medical food. The FDA states that medical foods are substances to be prescribed by a physician "for the specific dietary management of a disease or condition for which distinctive nutritional requirements, based on recognized scientific principles, are established by medical evaluation."¹⁴

As noted previously, evidence from past studies suggests that the individual components of LMF-MC-PP may address the underlying pathophysiology of DPN. Both pyridoxal 5'-phosphate and methylcobalamin monotherapy have been shown to be essential to critical peripheral nerve functions that are impaired in diabetic neuropathy.^{10,11,15} Also, 5-MTHF has been shown to improve endothelial function in patients with type 2 DM, possibly by enhancing synthesis of NO.^{8,9,16} (Figure 3). The data from our pilot study involving a combination of metabolically active L-methylfolate, methylcobalamin, and pyridoxal 5'-phosphate are consistent with studies of monotherapy with these B vitamins and suggest that LMF-MC-PP modifies the pathophysiology of DPN, renews perfusion of peripheral nerves, and improves sensory perception in patients with DPN.

Evidence suggests that the pathogenesis of DPN is multifactorial and involves the effects of hyperglycemia, oxidative stress, mitochondrial dysfunction, and impairment of transport of nutrients within nerve axons as a result of accumulation of advanced glycation end products.^{17,18} Oxidative stress in patients with diabetes is known to reduce endothelial production of NO, a gaseous free radical and important mediator of vascular smooth muscle relaxation and tissue repair.¹⁹ More specifically,

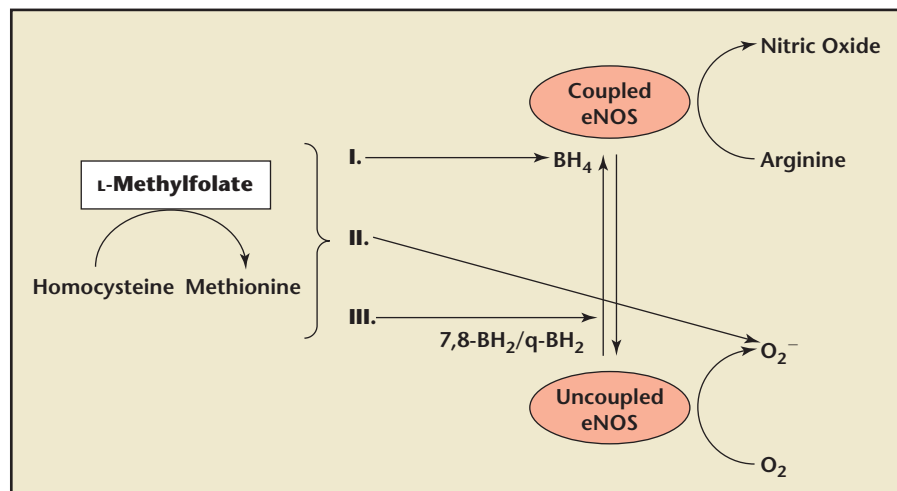


Figure 3. Nitric oxide synthesis and potential mechanisms of action of L-methylfolate. Diagram illustrates nitric oxide (NO) synthesis from L-arginine, as catalyzed by endothelial NO synthase (eNOS), and potential mechanisms for beneficial effects of L-methylfolate on eNOS. The beneficial effects of L-methylfolate may be explained by different mechanisms. I. Tetrahydrobiopterin (BH₄) rescue or BH₄ stabilization, in which L-methylfolate may stimulate endogenous BH₄ regeneration from quinoid-dihydrobiopterin (q-BH₂) or lead to chemical stabilization of BH₄. II. Antioxidant effects, in which L-methylfolate may act as a direct antioxidant. III. Direct effect on eNOS, in which L-methylfolate reduces superoxide generation and increases NO synthesis in a BH₄-dependent manner. Adapted with permission from Verhaar MC, Stroes E, Rabelink TJ. Folate and cardiovascular disease. *Arterioscler Thromb Vasc Biol.* 2002;22:6-11.⁹

oxidative stress "uncouples" endothelial NO synthase (eNOS), thereby reducing the availability of NO and promoting constriction of blood vessels.^{9,19,20} (Figure 3). Reduced capillary flow associated with this endothelial dysfunction is believed to cause nerve hypoxia, and, eventually, DPN.^{8,15} Reduced synthesis and bioactivity of NO also appear to be associated with impaired wound healing in patients with diabetes.^{21,22}

Treatment with oral 5-MTHF offers several potential advantages, as compared with folic acid; 5-MTHF has 7 times the biologic activity of folic acid. Also, unlike folic acid, 5-MTHF is metabolically active and does not undergo the complex 4-step conversion process required for the activation of folic acid. Furthermore, because up to 50% of the potential patient population has a genetic polymorphism that precludes complete conversion of folic acid to L-methylfolate, oral 5-MTHF offers

this group of patients the advantage of greater utilization.^{23,24}

The PSSD method used in this study to assess 1-point sensory (tactile) and 2-point (discriminatory) sensation is a quantitative sensory test. When used for 2-point testing, the PSSD measures paired neurosensory end-organ stimuli to the skin. During testing, 2 identical hemispherical metal prongs (each of which has its own force transducer) are placed against the skin. In the current study, we assessed static touch, which is mediated by the Merkel cell-neurite complex related to the slowly adapting, large, thinly myelinated group A-beta fibers (moving touch, which is mediated by mechanoreceptors called Meissner and Pacinian corpuscles, is transmitted by a quickly adapting group of A-beta fibers). During PSSD testing, the cutaneous pressure threshold that distinguishes 1 stimulus from 2 repetitive stimuli is calculated by a computer, according to the method of limits.

Because PSSD testing involves no needle sticks or electric shocks, patients have no pain and typically are willing to undergo testing more than once, if needed. Compared with other modalities of sensory testing, including Semmes-Weinstein monofilament and electromyography/nerve conduction velocity testing, the PSSD has greater sensitivity at detecting early sensory fiber loss.²⁵⁻³⁰ Because PSSD testing is an objective measure, it may be used to monitor neuronal response to treatment, as it was in this study.²⁹

The 2-point static PSSD test results in the current study are particularly interesting. The finding that all 2-point static values for heel and great toe improved significantly during all follow-up periods suggests that LMF-MC-PP promotes restoration of A-beta fibers and, thereby, improves the ability of patients with diabetic neuropathy to perceive insults to the skin. Because restoration of cutaneous nerve fibers ordinarily

is preceded by return of peripheral nerve fiber function, it is possible that treatment with LMF-MC-PP may reduce risk of infection, ulceration, and amputation among diabetic patients with peripheral neuropathy.

Limitations in the design and interpretation of the current study include its relatively small sample size, the method of patient selection, possible volunteer bias, lack of control group, lack of blinding, lack of determination of the optimal duration of chronic therapy, the minimally subjective component of neurosensory testing, and the potential for a placebo effect. Moreover, the study did not control for variation of duration or severity of type 2 DM among patients and the use of insulin and/or symptom-ameliorating drugs by some patients.

Conclusions

The results of this pilot study suggest that LMF-MC-PP restores cutaneous

sensitivity in patients with DPN, possibly by promoting regeneration of damaged nerve fibers through increased availability of NO. The LMF-MC-PP was well tolerated, and no patients reported adverse effects. Thus, LMF-MC-PP should be considered for the treatment of DPN and its complications in appropriate patients.

Randomized controlled trials involving a larger population of patients with neuropathy are underway to confirm the preliminary findings of our study. Future prospective randomized controlled trials exploring mechanism of action and efficacy in larger populations of patients with DPN could determine whether LMF-MC-PP is indeed a disease-modifying therapy for DPN. Based on the potential mechanisms of action of LMF-MC-PP, clinical investigation of the efficacy and safety of combination LMF-MC-PP in patients with chemotherapy-induced peripheral neuropathy should also be considered. ■

Main Points

- Diabetic peripheral neuropathy (DPN) affects up to 60% of patients with diabetes mellitus (DM) and is a leading cause of disability in the United States. The severity of DPN increases with duration of DM, patient age, and degree of glycemic dysregulation. The gravity of DPN is underscored by the fact that DM has reached epidemic proportions within the last quarter century in the United States.
- Commonalities among several pathologic processes associated with DPN have prompted research into new pharmacologic interventions; oxidative stress and nerve perfusion deficits have emerged as leading candidates in the pathogenesis of DPN.
- We hypothesized that twice daily oral administration of 3 mg of L-methylfolate, 2 mg of methylcobalamin, and 35 mg of pyridoxal 5'-phosphate (LMF-MC-PP) improves sensory perception, as measured by 1-point (tactile) and 2-point (sensory discrimination) tests, in symptomatic type 2 DM patients with established DPN.
- Following baseline Pressure-Specified Sensory Device™ (PSSD) testing, patients were treated with LMF-MC-PP twice daily for 4 weeks and then once daily for an additional 48 weeks. After 6 months and 1 year of treatment, patients underwent follow-up (PSSD) testing.
- Our study found that treatment with LMF-MC-PP was associated with statistically significant improvement in both 1- and 2-point static PSSD measurements in the great toe and heel at 6 months and 1 year.
- The data from our pilot study involving a combination of metabolically active L-methylfolate, methylcobalamin, and pyridoxal 5'-phosphate are consistent with studies of monotherapy with these B vitamins and suggest that LMF-MC-PP modifies the pathophysiology of DPN, renews perfusion of peripheral nerves, and improves sensory perception in patients with DPN.

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References

1. Frykberg RG, Lavery LA, Pham H, et al. Role of neuropathy and high foot pressures in diabetic foot ulceration. *Diabetes Care*. 1998;21:1714-1719.
2. National Diabetes Information Clearinghouse. National Diabetes Statistics, 2007. National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK). <http://diabetes.niddk.nih.gov/dm/pubs/statistics/#complications>. Accessed November 15, 2010.
3. Shun CT, Chang YC, Wu HP. Skin denervation in type 2 diabetes: correlations with diabetic duration and functional impairments. *Brain*. 2004;127:1593-1605.
4. Boulton AJM. Management of diabetic peripheral neuropathy. *Clin Diabetes*. 2005;23:9-15.
5. Argoff CE, Backonja MM, Belgrade MJ, et al. Consensus guidelines: treatment planning and options. Diabetic peripheral neuropathic pain. *Mayo Clin Proc*. 2006;81(4 suppl):S12-S25.
6. Frykberg RG, Zgoni T, Armstrong DG. Diabetic foot disorders: A clinical practice guideline (2006 revision). *J Foot Ankle Surg*. 2006;45(5 suppl):S1-S66.
7. Veves A, Backonja M, Malik RA. Painful diabetic neuropathy: epidemiology, natural history, early diagnosis, and treatment options. *Pain Med*. 2008;9:660-674.
8. van Etten RW, de Koning EJ, Verhaar MC, et al. Impaired NO-dependant vasodilation in patients with type II (non-insulin-dependent) diabetes mellitus is restored by an acute administration of folate. *Diabetologia*. 2002;45:1004-1010.
9. Verhaar MC, Stroes E, Rabelink TJ. Folate and cardiovascular disease. *Arterioscler Thromb Vasc Biol*. 2002;22:6-11.
10. Yaqub BA, Siddique A, Sulimani R. Effects of methylcobalamine on diabetic neuropathy. *Clin Neurol Neurosurg*. 1992;94:105-122.
11. Watanabe T, Kaji R, Oka N. Ultra-high dose methylcobalamin promotes nerve regeneration in experimental acrylamide neuropathy. *J Neurol Sci*. 1994;122:140-143.
12. McCann VJ, Davis RE. Serum pyridoxal concentration in patients with diabetic neuropathy. *Aust N Z J Med*. 1978;8:259-261.
13. Metanx [package insert]. Covington, LA: PamLab, LLC; 2008.
14. US Department of Health and Human Services, Food and Drug Administration, Center for Food Safety and Applied Nutrition. Guidance for Industry: Frequently Asked Questions About Medical Foods. <http://www.fda.gov/Food/GuidanceComplianceRegulatoryInformation/GuidanceDocuments/MedicalFoods/ucm054048.htm>. Accessed November 17, 2010.
15. Sheehan P. An overview of peripheral neuropathy in diabetes. *Vasc Dis Manag*. 2009;6:68-71.
16. Mangoni AA, Sherwood RA, Asonganyi B, et al. Short-term folic acid supplementation enhances endothelial function in patients with type 2 diabetes. *Am J Hypertens*. 2005;18:220-226.
17. Brownlee M. Biochemistry and molecular cell biology of diabetic complications. *Nature*. 2001;414:813-820.
18. King RH. The role of glycation in the pathogenesis of diabetic polyneuropathy. *Mol Pathol*. 2001;54:400-408.
19. Schwentker A, Billiar TR. Nitric oxide and wound repair. *Surg Clin North Am*. 2003;83:521-530.
20. Chen Z, Karaplis AC, Ackerman SL. Mice deficient in methylenetetrahydrofolate reductase exhibit hyperhomocysteinemia and decreased methylation capacity, with neuropathology and aortic lipid deposition. *Hum Mol Genet*. 2001;10:433-443.
21. Veves A, Akbari CM, Primavera J, et al. Endothelial dysfunction and the expression of endothelial nitric oxide synthase in diabetic neuropathy, vascular disease, and for ulcerations. *Diabetes*. 1998;47:457-463.
22. Boykin JV Jr, Baylis C. Homocysteine: a stealth mediator of impaired wound healing: a preliminary study. *Wound*. 2006;18:1-12.
23. Willems FE, Boers GH, Blom HJ, et al. Pharmacokinetic study on the utilisation of 5-methyltetrahydrofolate and folic acid in patients with coronary artery disease. *Br J Pharmacol*. 2004;141:825-830.
24. Venn BJ, Green TJ, Moser R, Mann JI. Comparison of the effect of low-dose supplementation with L-5 methyltetrahydrofolate or folic acid on plasma homocysteine: a randomized placebo-controlled study. *Am J Clin Nutr*. 2003;77:658-662.
25. Dellon ES, Crone S, Mourey R, Dellon L. Comparison of the Semmes-Weinstein monofilament with the pressure-specifying sensory device. *Restor Neurol Neurosci*. 1993;5:323-326.
26. Tassler PL, Dellon AL, Scheffler NM. Computer-assisted measurement in diabetic patients with and without foot ulceration. *J Am Podiatr Med Assoc*. 1995;85:679-684.
27. Barber MA, Conolley J, Spaulding CM, Dellon AL. Evaluation of pressure threshold prior to foot ulceration: one- versus two-point static touch. *J Am Podiatr Med Assoc*. 2001;91:508-514.
28. Weber RA, Schuchmann JA, Albers JH, Ortiz J. A prospective blinded evaluation of nerve conduction velocity and the Pressure-Specified Sensory Testing in carpal tunnel syndrome. *Ann Plast Surg*. 2000;45:252-257.
29. Valdivia JM, Dellon AL, Weinand ME, Maloney CT Jr. Surgical treatment of peripheral neuropathy: Outcomes from 100 consecutive decompressions. *J Am Podiatr Med Assoc*. 2005;95:451-454.
30. Wood WA, Wood MA, Werter SA, et al. Testing for loss of protective sensation in patients with foot ulceration: A cross-sectional study. *J Am Podiatr Med Assoc*. 2005;95:469-474.