

Vestibulodynia: Synergy Between Palmitoylethanolamide + Transpolydatin and Transcutaneous Electrical Nerve Stimulation

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■ Abstract

Objective. The study aimed to assess the effect of palmitoylethanolamide + transpolydatin combination in patients with vestibulodynia undergoing transcutaneous electrical nerve stimulation (TENS) therapy and to confirm the effectiveness of TENS also in a domiciliary protocol. The study is based on the premise that palmitoylethanolamide + transpolydatin combination may contribute to a down-regulation of mast cell hyperactivity, which is believed to be responsible for the proliferation and sprouting of vestibular pain fibers and the associated hyperalgesia and allodynia.

Materials and Methods. Twenty women with vestibulodynia were randomly assigned to receive oral palmitoylethanolamide (PEA) 400 mg and transpolydatin 40 mg or placebo, twice daily for 60 days. All patients underwent TENS therapy in a self-administered home protocol. Visual analogue scale (VAS), Marinoff score for dyspareunia, and current perception threshold obtained from the vulvar vestibule were assessed at baseline and at the end of treatment.

Results. The patients received a mean of 26.7 TENS sessions. All scores in the 2 groups improved significantly, although the level of improvement was similar between the groups (VAS, $p < .57$; dyspareunia, $p < .38$). Nevertheless, the analysis of regression of symptoms related to the duration of disease revealed the therapy to be more effective when PEA + transpolydatin is included in cases with more recent disease onset, as compared with the placebo group

(PEA: VAS, $p < .01$; dyspareunia, $p < .01$) (placebo: VAS, $p =$ nonsignificant; dyspareunia, $p =$ nonsignificant).

Conclusions. This study confirms that TENS is of significant benefit in the management of vestibulodynia, also in a home environment. PEA + transpolydatin can be a value-added treatment adjunct when the onset of vestibulodynia is more recent or when the disease relapses. ■

Key Words: vulvodynia, transcutaneous electrical nerve stimulation, vulvar vestibulitis syndrome, palmitoylethanolamide, transpolydatin

Vulvodynia is a common vulvar discomfort, most often described as a burning pain, occurring in the absence of relevant visible findings or a specific, clinically identifiable disorder [1]. The predominant form of vulvodynia, provoked vestibulodynia previously known as vulvar vestibulitis, is characterized by burning and cutting pain localized to the vulvar vestibule in response to light touch [2]. Introital dyspareunia, the intensity of which may inhibit or prevent intercourse, is often the presenting symptom. Pain may occur in other situations where pressure is exerted upon the vestibule, such as tampon insertion or bike riding, walking, and wearing tight clothing, or prolonged sitting.

The etiology of vulvodynia is not fully understood. Two findings suggest that neuropathic mechanisms may underlie these clinical symptoms where neurogenic inflammation is involved as follows: the presence of

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more activated mast cells and an increased density of free nerve endings in the vestibular mucosa in cases of provoked vestibulodynia [3–5]. Brain imaging of patients with provoked vestibulodynia reveals patterns of activity similar to those observed in experimental and clinical pain, as well as neuroanatomic abnormalities suggestive of compensatory central reorganization secondary to chronic pain [6]. Numerous medications and clinical approaches have been reported as efficacious, mostly in uncontrolled trials, but none have been recognized as optimal [7].

In light of the complex neuropathology of this syndrome, an effective therapeutic approach should target both peripheral and central neural sensitization. Transcutaneous electrical nerve stimulation (TENS) was demonstrated to be effective in patients with provoked vestibulodynia [8, 9]. TENS is a technique that provides neuromodulation (peripheral nociceptive blockade and central inhibition) via an electrical stimulus.

Mast cells are the main source of inflammatory mediators (nerve growth factor, tumor necrosis factor α , proteases, and cytokines) implicated in local tissue inflammation, in proliferation and sprouting of pain fibers, with hyperalgesia and allodynia, and in neurogenic inflammation, which is characterized by a progressive shift from nociceptive to neuropathic pain [10].

Patients with provoked vestibulodynia have increased numbers of mast cells near pain fibers as well as degranulated vulvar vestibule mast cells. Palmitoylethanolamide (PEA), a fatty acid ethanolamide congener of the endocannabinoid anandamide [11], reduces mast cell activation and the subsequent release of bioactive mediators. Our premise is that PEA may contribute to a down-regulation of hyperactivated mast cells responsible for the proliferation and sprouting of vestibular pain fibers [12].

The present study has 2 objectives as follows: (1) to assess the effect of PEA + transpolydatin in patients with vestibulodynia undergoing TENS therapy as a multimodal treatment strategy; and (2) to confirm the effectiveness of TENS also in a domiciliary protocol.

MATERIALS AND METHODS

Twenty women entered the study. All were diagnosed with vestibulodynia due to the coexistence of the following conditions: a history of at least 6 months of vulvar pain upon tampon insertion or attempted intercourse, and a positive cotton swab test result, that is, tenderness at palpation of the vestibular area with a cotton tip applicator [13], in the absence of other causes for these

findings. The study was a randomized, double-blind, placebo-controlled trial. Randomization was determined by a computer-generated number list. Institutional review board approval for the study was obtained, and all participating individuals gave written informed consent.

At the first assessment, symptoms of irritation and burning were evaluated on a 10-cm visual analogue scale (VAS). Dyspareunia was recorded and graded on a 0 to 3 score according to the Marinoff dyspareunia scale [14]. Patients also underwent current perception threshold (CPT) testing, a technique which quantifies the functional integrity of specific afferent nerve fibers from the periphery to the central nervous system. The CPT values were measured using the neurometer CPT/C electro diagnostic neurostimulator (Neurotron, Inc., Baltimore, MD), which emits sinusoid waveform constant alternating current stimuli at frequencies of 2,000 Hz (specific for large, myelinated A β fibers), 250 Hz (specific for A δ fibers) and 5 Hz (specific for C fibers), at intensity levels from 0.001 to 9.99 mA [15].

Vulvar vestibule CPT values (1 = 0.01 mA) were determined using a G-trode Vaginal/Rectal Electrode (Neurotron Inc, Baltimore, MD). At each frequency, stimulus intensity was incremented from 0 to a maximum of 9.99 mA until the patient was able to detect a sensation around the site of the electrode and was re-presented at decreasing intensities until it was no longer detected within a range of 0.10 mA. The patient was then presented with a series of choice tests consisting of randomly generated pairs of real and false (placebo) stimuli along with pairs of false stimuli, with each stimulus separated by a rest period, the threshold being determined after a minimum of 7 consecutive consistent forced-choice presentation responses. The CPT value was determined as an average of both the stimulus consistently detected and the stimulus that was not detected consistently.

Before randomization, patients were asked to stop any topical or systemic therapy they were taking. Patients were randomly assigned to receive oral PEA 400 mg and polydatin 40 mg or placebo, twice daily for 60 days. All patients received TENS therapy in a self-administered domiciliary protocol. A dual channel portable TENS unit (NeuroTrac Continence; Verity Medical, London, UK) was used, which produces a symmetrical biphasic wave and has 3 customizable mode programs. The stimulation was delivered through a commercially available plastic vaginal probe (Periprobe VAG2ST Beac, Pavia, Italy), 20 mm in diameter and 110 mm in length, with 2 gold metallic transversal rings as electrodes. It was inserted into the vagina for 20 mm.

Two customized programs were set according to our previous study [8]. The standard protocol for TENS was 15 minutes of 10-Hz frequency and pulse duration of 50 microseconds (first program), followed by 15 minutes of 50-Hz frequency and pulse duration of 100 microseconds (second program). All patients received a supervised TENS trial of TENS before use at home. The trial consisted in 6 to 7 sessions and served to familiarize the patient on use of TENS, while allowing the therapist to check that the patient was using the device properly.

In the TENS treatment protocol, the pulse is increased rapidly until the patient reports the onset of any sensation under the electrodes. The intensity is then increased slowly until this sensation reaches a level described as the maximum tolerable, without experiencing pain.

After completing the trial, the patient is consigned their TENS unit after verbal and written instruction, with a recommendation to perform home treatment 3 times each week.

Their clinical conditions were assessed by the VAS symptoms assessment, the Marinoff dyspareunia score, and the CPT values immediately after completing 2 months of treatment.

None of the patients received any other therapy during the TENS treatment period.

The characteristics of the study population are summarized in Table 1. Women in the PEA + polydatin and placebo groups were similar in age, parity, and symptoms at recruitment into the study; moreover, the CPT values were not statistically different between the 2 groups.

All eligible women were accepted to participate in the study, and no dropouts were recorded.

The EPI-INFO version 6.0 (Centers for Disease Control and Prevention, Atlanta, GA) was used for all statistical analyses. Descriptive statistical analysis (means,

Table 2. Posttreatment Scores of VAS and Marinoff Dyspareunia Scale in the 2 Groups

	PEA + polydatin group	Placebo group	<i>p</i>
VAS	2.2 (1.6)	2.3 (1.5)	NS
Marinoff dyspareunia scale	1.0 (0.9)	1.1 (0.9)	NS

Values are expressed means (1 SD). *p* values indicate comparisons with pretreatment scores.

PEA, palmitoylethanolamide; VAS, visual analogue scale; NS, not significant.

SDs, and percentages) was performed using Student *t* test and analysis of variance. Significance is taken at *p* < .05.

RESULTS

Patients received a mean of 26.7 (range = 21–35) TENS sessions. All scores in the 2 groups improved significantly by the end of treatment compared with prestudy values (see Table 2). The results reported here are consistent with our recent randomized, controlled trial demonstrating that TENS is an effective treatment of vestibulodynia [8]. However, none of the observed differences were statistically significant between the PEA and placebo groups.

To further evaluate patient responses, results were analyzed with respect to factors other than symptoms during the study period. An analysis of regression of symptoms related to the duration of disease revealed that TENS was more effective in those cases where the disease is more recent and PEA + polydatin used as an add-on therapy (PEA + polydatin: VAS, *p* < .01; dyspareunia, *p* < .01) (placebo: VAS, *p* = nonsignificant; dyspareunia, *p* = nonsignificant). In the latter, statistical analysis indicated significance, with correlation coefficients of 0.679 and 0.747 for VAS and dyspareunia, respectively. Significance of the regression was *p* < .01.

Table 1. Characteristics of the Study Population.

	PEA +polydatin group	Placebo group	<i>p</i>
Age, mean (range) y	34.4 (18–48)	31.8 (23–47)	NS
Nulliparous, <i>n</i> (%)	10/10 (100)	9/10 (90)	NS
Duration of symptoms, mean (range), mo	27.5 (6–80)	34.2 (12–72)	NS
Sexually active, <i>n</i> (%)	7/10 (70)	8/10 (80)	NS
VAS, mean (SD)	5.8 (1.1)	6.2 (1.1)	NS
Marinoff dyspareunia scale, mean (SD)	2.8 (0.4)	2.6 (0.5)	

PEA, palmitoylethanolamide; NS, not significant; VAS, visual analogue scale.

Table 3. Results of CPT Measurement (100 = 1.0 mA) at 3 Selected Stimulation Frequencies Before and After Therapy

Subject group		2,000 Hz (Aβ fibers)	250 Hz (Aδ fibers)	5 Hz (C fibers)
PEA + polydatin	Basal	541.9	256.9	82.5
	After therapy	575	259.2	139.5
	Difference, %	5.7	0.8	40.8
Placebo	Basal	598.5	214	99.7
	After therapy	558.8	217.8	104.5
	Difference, %	–6.6	1.7	4.5

Data are expressed as means (Hz = cycles per second). CPT, current perception threshold; PEA, palmitoylethanolamide.

Mean CPT values are reported in Table 2. Women in the PEA + polydatin group showed mean CPT values at a 5-Hz stimulation (C fibers) 10-fold lower compared with placebo (reduction of 40% vs 4.59%), whereas the CPT values at 2,000-Hz (A β) showed a less significant reduction than values at 5-Hz stimulation (12.8%). The 250-Hz stimulation (A δ fibers) values showed a similar reduction among the 2 groups (0.8% vs 1.7%) (see Table 3).

The overall incidence of adverse events was very low, and none led to treatment discontinuation. Adverse events were reported in 3 patients, 2 in the PEA + polydatin group and 1 in the placebo group (mild and transient gastrointestinal symptoms, none leading to withdrawal and dropout).

DISCUSSION

This study confirms that TENS is of significant benefit in the management of vestibulodynia [8, 9], also when used at home. An increase in intraepithelial innervation and in the number of C-afferent nociceptors has been demonstrated in biopsy specimens of the vestibular mucosa obtained from women with vulvodynia [3, 4]. An increase in the number of active mast cells has also been shown histologically in vestibulodynia patients [5, 16]. Activated mast cells secrete a wide array of pluripotent inflammatory mediators, including nerve growth factor, proteases, cytokines (especially interleukin 1 β [IL-1 β]), and tryptase, which can sensitize C-nerve fibers and induce their proliferation [10].

Vestibulodynia shares features common to neuropathic pain, including allodynia and hyperalgesia. Histological correlates include the following: (1) a significant increase of proliferating nerve fibers (which is the morphological and anatomic correlate and first contributor to the hyperalgesia reported by patients); (2) a superficialization of pain fibers that cross the epidermal and mucous basal membrane along the “tunnel” created by tryptase and heparanase. The latter is likely to be the anatomic correlate of allodynia, that is, the shift from a perception of the tactile stimulus to a burning one.

The present findings do not support previous claims of an absence of focal pathological finding. The shift to neuropathic pain involves complex changes in the central nervous system, which include hyperactivation of spinal cord microglia and neuronal reorganization in the thalamic, limbic, and cortical sensory areas [11]. Central nervous system involvement suggests that key steps of vulvodynia may well lie in the central processing of painful sensory inputs from the vulva.

Treatment strategies for vestibulodynia should thus address the multifactorial nature of neuropathic pain, including the multiple levels of pain mechanisms [17].

The previous information provided the rationale for the current study, based on the use of agents capable of controlling mast cell activator release. The latter include a class of molecules termed *ALIAmides* (from the acronym Autacoid Local Injury Antagonism), naturally occurring lipid amides derived from membrane fatty acids and structurally related to endocannabinoids [18]. PEA is considered the prime example of an *ALIamide*: it negatively modulates mast cell activation and down-regulates their inflammatory activity [12], while also acting as a ligand for peroxisome proliferator-activated receptor α and the transient receptor potential vanilloid type 1 channel—actions that likely participate in its analgesic activity [18].

Polydatin, a compound that decreases levels of the proinflammatory cytokine IL-17 levels, was used in association with PEA [19].

Inflammation is now recognized as a critical aspect in the development of neuropathic pain, although its role in vestibulodynia is debated. Although there is histological evidence for an inflammatory process (increased mast cell number, presence of degranulated mast cells, and mast cells near pain nerve fibers) [4, 5], others have failed to find active tissue inflammation in vestibulodynia patients, as assessed by up-regulated expression of inflammatory markers (cyclooxygenase 2 and inducible nitric oxide synthase) [20]. Reasons for these differences may include heterogeneity of inflammatory factors/agents (infection, mechanical trauma, chemical agents, physical lesions, neurogenic inflammation) that trigger vestibular tissue reaction and the lag time from onset of symptoms. A “neurogenic inflammation” may further contribute to pain when prolonged, or repeated “trigger” inflammatory events such as infections, mechanical trauma, and repeated exposure to an irritant or allergen, maintain the anatomic vestibular inflammation with a subsequent increase in the number of activated mast cells.

Recurrent *Candida albicans* is the most consistently reported finding associated with onset of vestibulodynia [21], and a reduced capacity to control infection due to polymorphisms in the genes coding for mannose-binding lectin or IL-1 β may be predisposing factors associated with vestibulodynia [22, 23]. Similar mechanisms may occur in cases of bladder pain syndrome/interstitial cystitis, a condition that may occur simultaneously with vestibulodynia [24].

Vulnerability to vestibulodynia may be reduced also by agents, which prevent mast cell activation. The results reported here demonstrate a synergic action of TENS in combination with PEA + polydatin in patients with a more recent disease onset.

Mast cell mediator release in inflamed tissue changes over time [10]. In early stage of disease (acute inflammatory response), mast cell numbers are significantly increased in the vestibular mucosa; however, as inflammation becomes more chronic, the number of mast cells decreases and C-afferent nociceptors in the vestibular mucosa proliferate [25]. At this late stage of the inflammatory process, neuropathic pain symptoms, such as hyperalgesia and allodynia, became prominent.

An interesting observation was the marked difference in CPT values for C fibers (which are mainly responsible for pain perception) in the PEA + polydatin versus control groups (40% and 4.59% reduction, respectively). Conceivably, this could reflect a synergic action between TENS and PEA + polydatin on inflammation-triggered peripheral sensitization of sensory nerves in the vestibular tissue.

The less consistent modification of CPT values of A β fibers may result from a slow “switch off” of the overactive malfunctioning pain sensory system, mainly linked to the TENS, in which PEA + polydatin has a preventive effect on mast cells reactivation. However, type A β fibers can become involved in pain sensation (allodynia), resulting from prolonged and intensified nociceptive C fiber activity which sensitizes sensory neurons to low-threshold A β fiber activity.

The mode of action of TENS is complex with 2 mechanisms postulated as follows: (1) a block of information traveling along nociceptive fibers via stimulation of large diameter afferent A β fibers, thereby inhibiting the small nociceptive fibers A δ and C by activating inhibitory interneurons in the spinal cord dorsal horn [26]; (2) release of endogenous opioids by stimulation of small diameter afferent and motor fibers. Pulse durations of 30 to 100 microseconds activate large-diameter fibers but not smaller nociceptive fibers, whereas pulses of 100 to 200 microseconds simultaneously stimulate “sensitive” and “pain” fibers. In this case, analgesic action results from activation of descending pain inhibitory pathways [27]. This study demonstrates the therapeutic feasibility of TENS also in a home setting. TENS is noninvasive, inexpensive, safe, and easy to use.

Identifying an effective therapy for a patient with vestibulodynia can be quite time-consuming. Some treatments, such as amitriptyline, produce systemic adverse ef-

fects such as drowsiness, which may limit their use [28]. Other effective therapies, for example, biofeedback of the pelvic floor musculature, can require a large-time commitment [29].

Our multimodal approach (TENS + PEA + polydatin) demonstrated effectiveness in a relatively short time (2 months) with an overall low incidence of adverse events, none of which led to treatment discontinuation. This double-blind, placebo-controlled trial is the first to provide measurements of subjective and objective components of patient responses using CPT evaluation of vestibular nerve fibers. Few treatment modalities currently in use have been subject to controlled evaluation of efficacy: TENS is one of them [8].

In summary, the findings reported here propose the use of PEA + polydatin as a coadjuvant therapy in patients with vestibulodynia, especially in cases of recent disease onset. Future trials need to be carried out to define timing for optimum efficacy of PEA + polydatin treatment. Moreover, trials with larger patient numbers are needed to confirm our observations and to understand which patients are more likely to benefit from this treatment regimen.

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