Minimally Invasive Therapy Using Intralesional OnabotulinumtoxinA in Peyronie's Disease

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Purpose: To determine the effectiveness of intralesional administration of onabotulinumtoxinA in patients with Peyronie's disease (PD).

Materials and Methods: A prospective therapeutic cohort study was undertaken in patients aged ≥ 18 years with stable PD. Intervention included one-time intralesional application of 100 U of onabotulinumtoxinA. We included 22 patients who attended the urology clinic from October 1, 2011 to June 30, 2012. Primary outcome measure was degree of curvature. Secondary outcome measures were thickness of the fibrous plaque, improvement in erectile function and pain. Erectile function was evaluated using the International Index of Erectile Function (IIEF-5) questionnaire. The Visual Analog Scale (VAS) was used to measure pain during an erection. Statistical analyses were performed by Pearson's chi-squared test for categorical variables and student's *t*-test for quantitative variables. Any *P* value < .05 was considered statistically significant.

Results: The size of the fibrous plaque was reduced from 0.34 ± 0.20 to 0.27 ± 0.13 cm after treatment (*P* = .014). The curvature initially averaged 32.95 ± 9.21°, and improved to 25 ± 9.38° (*P* = .025). According to the Kelami classification, the curvature was < 30° in 14 cases (63.6%) and was 30°-60° in eight cases (36.4%). At 16 weeks, the curvature was < 30° in 19 cases (86.4%) and 30°-60° in three cases (13.6%). The IIEF-5 score was 16.18 ± 4.46 before treatment and 18.22 ± 4.55 after treatment (*P* = .002). Pain was reduced from 3.36 ± 3.48 before treatment to 1.14 ± 1.58 after treatment (*P* = .001).

Conclusion: The administration of onabotulinumtoxinA may improve the clinical manifestations of PD resulting from fibrosis, thus improving sexual function in patients.

Keywords: penile induration; therapy; treatment outcome; drug therapy; botulinum toxins; type A acetylcholine release inhibitors.

INTRODUCTION

Peyronie's disease (PD) involves scarring and fibrosis of connective tissue of the tunica albuginea via chemical tactile mediators. Peyronie's fibrous plaque causes pain during erection and, therefore, sexual dysfunction. The disease may also include penile deformity, sometimes > 45°, causing distal flaccidity and rendering coitus impossible.[1,2] The management of this disease is controversial and there is no gold standard treatment.

OnabotulinumtoxinA has been shown to reduce fibrosis in hypertrophic scars and keloids,[3-6] thus offering new avenues for the study and treatment of illnesses such as PD. However, recent evidence does not support a beneficial effect of botulinum toxin in the treatment of keloid scars.[7,8] Yet, the toxin is used widely in the treatment of various urological diseases. We decided to investigate any beneficial effect of one intralesional administration of the onabotulinumtoxinA to evaluate the clinical effectiveness in patients with PD.

MATERIALS AND METHODS

Study Population

A prospective cohort for therapeutic intervention was designed using 22 patients seeking consultation for sexual intercourse problems related to PD at the Department of Urology of the High Specialty Medical Unit, Specialties Hospital of the Western Medical Center, Mexican Institute of Social Security in Guadalajara, Mexico from October 1, 2011 to June 30, 2012. The inclusion criteria were patients aged ≥ 18 years with PD confirmed by the presence of stable plaque (at least 12 months from the onset of illness) as detected using ultrasonography; no medical or surgical treatment for the previous 6 months; and authorization obtained through signed informed consent. Patients were excluded if they were in an active stage, had calcified plaques, or had a history of allergy or sensitivity to any of the components of the medication (onabotulinumtoxinA). The study variables were clinical history, degree of curvature, pain, erectile dysfunction

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Table 1. Demographic and clinical characteristics of study subjects.

Variables	No.	%
Comorbidity		
Diabetes mellitus	7	31.8
Hypertension	13	59.1
Cardiac disease	3	13.6
Spinal surgery	3	13.6
Hypercholesterolemia	4	18.2
Hypertriglyceridemia	5	22.7
Genital trauma	8	36.3
Previous oral treatments		
Colchicine	9	40.9
Vitamin E	7	31.8
Nonsteroidal anti-inflammatory drugs	3	13.6
Pentoxifylline	1	4.5
Previous intralesional treatments		
Verapamil	2	9
Interferon	1	4.5

(ED) and thickness of the fibrous plaque. The follow-up was for 16 weeks, with visits performed at 2, 4, 8, and 16 weeks. Measurements were taken at the beginning and at 16 weeks after treatment.

The primary outcome measure was the degree of curvature. The secondary outcome measures were the thickness of the fibrous plaque and improvement in ED and pain during erection. The penis was assessed on its ventral surface via high-frequency lineal tests, together with longitudinal and cross-sectional views. The penile curvature was measured (at the baseline and at the follow-up) during penile erection after the administration of intracavernosal onabotulinumtoxinA, and measurements were recorded by a single person using a goniometer at the point of maximum angulation during maximum penile rigidity. The severity of deformity, which was assessed using the Kelami modified classification, was divided into three types: mild penile deformity (≤ 30°), moderate penile deformity (31°-60°), and severe penile deformity (> 60°).[8] The Visual Analog Scale (VAS) was used to measure pain during an erection; the pain was graded on a 0–10 scale represented on a 10 cm line, with 0 as no pain and 10 as maximum pain.

Doppler ultrasonography, with a 7.5 to 12 MHz linear transducer for small organs and superficial lesions, was used to determine the thickness of the plaque, as well as its size, depth, and localization (dorsal, left lateral, right lateral, ventral, dorsoventral, or ventrolateral) and the presence of hyperechogenicity suggestive of calcification. Erectile function was evaluated using the International Index of Erectile Function (IIEF) questionnaire, with normal erectile function as 22-25 points, mild dysfunction 17-21, mild-to-moderate ED 12-16, moderate ED 8-11, and severe ED 5-7 points.

OnabotulinumtoxinA Administration Technique

To prepare the vials, dilution was performed using a sterile technique by introducing 4 mL of solution slowly into 100 U of onabotulinumtoxinA without forming bubbles. After the penile asepsis, the solution was injected into the fibrous plaque in quadrants, using a 20-gauge needle.

Statistical Analysis

A descriptive analysis was performed using raw numbers, percentages, and measures of central tendency and spread. The analytic phase was carried out using Pearson's chi-squared test for categorical variables and student's *t*-test for quantitative variables. Statistical significance was set at $P < .05$, and all tests were two-sided. Statistical Package for the Social Science (SPSS Inc, Chicago, Illinois, USA) version 17.0 was used for statistical analyses.

Ethical Considerations

The study was conducted according to the principles of the 1989 Declaration of Helsinki and the Mexican Health Guidelines. The Ethics and Research Committees of the Specialties Hospital approved all protocols (code 2012/1301/78). Full written informed consent was obtained from all patients before their inclusion in the study.

RESULTS

The average age was 56.6 ± 10.1 years (range, 35-68 years). The associated morbid conditions are described in **Table 1**. The history of genital trauma was present in eight cases. At the onset of the disease, during the active phase, the first symptom was pain in 10 (45.5%) patients, palpable fibrous plaque in 5 (22.7%) and penile deformity in 7 (31.8%) patients (**Table 2**). Three (13.64%) patients mentioned a sudden onset of the disease; in 19 (86.36%) patients the progression was gradual and then stable. The plaque fibrosis was progressive in 17 (77.27%) patients,

Table 2. Clinical characteristics of patients with Peyronie's disease before treatment with onabotulinumtoxinA.

First Sign or Symptom	N	%
Pain during erection	10	45.5
Plaque	5	22.7
Penile curvature	7	31.8
Pain during intercourse	12	54.5
Penile deformity		
Sudden	3	13.6
Gradual	19	86.4
Sexual trauma	6	27.3
Other	2	9.1
Pain		
Initial	5	22.7
Always	12	54.5
Absent	5	22.7
Penile curvature direction		
Dorsal	8	36.4
Ventral	5	22.7
Left lateral	3	13.6
Right lateral	6	27.3

Table 3. International Index of Erectile Function (IIEF-5) score before and after the administration of onabotulinumtoxinA.

Initial	No.	%	Final	No.	%
22–25	2	9.1	22–25	7	31.8
17–21	9	40.9	17–21	9	40.9
12–16	8	36.4	12–16	4	18.2
8–11	2	9.1	8–11	1	4.5
5–7	1	4.5	5–7	1	4.5

whereas 5 (22.73%) patients always had the same characteristics.

The penile curvature had a dorsal direction in 8 (36.4%) patients, ventral direction in 5 (22.7%) patients, left lateral direction in 3 (13.6%) patients, and right lateral direction in 6 (27.3%) patients. The penile deformities caused by the Peyronie's fibrous plaque included 4 (18.2%) cases of hinge or notch on the glans, 1 (4.5%) case of a hinge or notch at the base of the glans, 7 (31.8%) cases of curves of the erection to the right, 3 (13.6%) cases of curves of the erection to the left, 1 (4.5%) case of hourglass-shaped plaque in the base, 1 (4.5%) case of hourglass-shaped plaque in the middle of the penis and 1 (4.5%) case of hourglass-shaped plaque close to the glans. The penile curvature was evident in 4 (18.2%) patients in flaccid state.

Pain during sexual intercourse was reported by 5 (22.7%) patients at the beginning of the illness, 12 (54.5%) had pain throughout the duration of the illness, and 5 (22.7%) patients never had pain. Thirteen (59.1%) patients reported dyspareunia in their partners, 20 (90.9%) reported difficulty in penetration due to the curvature, 4 (18.2%) patients had a "hinge" effect and 11 (50%) patients reported loss of firmness. During the progression of the disease, all patients reported shortening of the penis, by an average of 2.6 ± 1.54 cm. The relationships of the couples were affected in 13 (59.1%) patients, and emotional state was affected in 16 (72.7%) patients. Libido was normal in 13 (59.1%), reduced in 6 (27.3%) and increased in 3 (13.6%) patients. The ability to ejaculate was preserved in 21 (95.5%) patients; coitus was done by 20 (90.9%), masturbation was performed by 8 (36.4%) and oral sex was used by 2 (9.1%) patients. The localization of the fibrous plaque, as assessed by ultrasonography, had the following distribution: dorsal in 7 (31.8%), ventral in 1 (4.5%), dorsolateral in 13 (59.1%) and ventrolateral in 1 (4.5%) patients. No complications were observed after intralesional application of onabotulinumtoxinA. The dimensions of the penis were similar before and after the administration of the drug, with an average of 10.34 ± 0.96 cm.

Table 4. Response to the intralesional administration of onabotulinumtoxinA.

Variables	Initial	Final	*P* Value
Penile curvature	32.95 ± 9.21	25 ± 9.38	.025
Plaque thickness	0.34 ± 0.20	0.27 ± 0.13	.014
IIEF-5 score	16.18 ± 4.46	18.22 ± 4.55	.002
VAS score	3.36 ± 3.48	1.14 ± 1.58	.001

Abbreviations: IIFE, International Index of Erectile Function; VAS, Visual Analog Scale.

The penile curvature (according to the Kelami classification) was 32.95° ± 9.21° before treatment and 25° ± 9.38° after treatment (P = .025). Nineteen (86.4%) patients had mild-to-moderate ED, and 1 (4.5%) patient had severe ED. After the treatment, 7 (30.8%) patients had normal sexual function, 14 (63.6%) patients still had mild-to-moderate ED and only 1 (4.5%) patient had severe ED. **Table 3** lists these characteristics with statistical differences after the intralesional application of the toxin (P = .002).

The average thickness of the initial fibrous plaque was 0.34 ± 0.20 cm; this was reduced to 0.27 ± 0.13 cm after the treatment (P = .014). The penile angle before treatment was 32.95° ± 9.21°; after treatment, it was reduced to 25° ± 9.38° (P = .025). The total IIEF-5 score was 16.18 ± 4.46 initially and 18.22 ± 4.55 after treatment (P = .002). There was also a significant reduction in pain, from 3.36 ± 3.48 to 1.14 ± 1.58 (P = .001) on the VAS, as shown in **Table 4**.

DISCUSSION

The pharmacological and surgical treatments that have been used for decades to manage PD include oral, topical and local therapies, as well as extracorporeal shock wave lithotripsy and intralesional treatments. The results vary, as the illness has a chronic progression. Patients' quality of life decreases in relation to their own and their partners' sexual activity.[9] Social taboos and the desire to maintain a masculine façade hinder the seeking of treatment in the early stages of the disease. Primary care providers often have little knowledge of the illness, and patients lack interest in follow-up because of the low efficacy of the treatment. Thus, this disease is chronic, progressive and without a definite treatment.[10-12]

Trost and colleagues published the results of a retrospective series of 127 patients who received intralesional interferon-α2b. They reported improvement in 54% of the patients, and a reduction of 9° in the curvature, but no effect on the IIEF score.[13] Intralesional and topical steroids have also been used to treat individuals under 50 years of age.[14] Hellstrom and colleagues showed that, in 31 of 50 patients treated with intralesional interferon α-2b (5 × 106 U in 10 mL of saline solution administered twice weekly for 12 weeks), the initial average curvature of 49.9° ± 2.4° was reduced to 36.4° ± 2.1° (P = .001) at the end of treatment period.[15] In 1980, Williams and Green reported the results of a consecutive series of patients who were treated with 25 mg of triamcinolone.[16] They observed a marked improvement in symptoms in patients with small and distal plaques. More recently, Dickstein and colleagues reported a small series of patients who received 50 mg of subcutaneous triamcinolone in the plaque area.[17] They observed a complete reduction in pain in the 16 cases, but no change in the curvature or the size of the plaque.

Collagenase from clostridium species was used by Jordan.[18] In 25 patients with repeated low-dose administrations, a 25% improvement in penile curvature was achieved in 57% of the patients; however, the majority of them had adverse effects such as pain, edema and ecchymosis. Orgotein, an anti-inflammatory drug with pronounced activity on superoxide dismutase, has been used in small groups of patients with PD and led to a reduction in plaque size and penile curvature.[19] However, patients experienced significant adverse events such as pain,

edema, paresthesia, dysesthesias and cutaneous rash. Calcium channel blockers (principally verapamil and nicardipine) have also been used for the treatment of PD. These drugs inhibit the formation of the extracellular matrix by reducing the proliferation of fibroblasts and increasing the amount of local collagenase.[20] In 1994, Levine and colleagues published the first series of 16 patients treated with multiple intralesional injections of verapamil.[21] A publication that followed included a larger number of patients (140 patients).[22] Levine and colleagues observed a significant reduction in plaque consistency and in penile curvature.

In the first clinical trial with verapamil administered by the intralesional application of saline solution, Rehman and colleagues demonstrated a 57% reduction in plaque size, with an initial average curvature of 37.7° ± 9.3° and a final result of 29.57° ± 7.3°.[23] However, the difference was not statistically significant. The plaque was softer in all patients, with a 42.8% erectile function improvement in the verapamil group. No systemic or local toxicity was reported.

Bennett and colleagues reported the prevention of the progression of PD using 6 intralesional injections of verapamil every 2 weeks (10 mg in 5 mL of saline solution), with a follow-up at 3 months after the last administration.[24] The study included 94 patients with an average age of 44 ± 18 years. Before the administration of the drug, 86% of the patients had dorsal penile curvature and 14% had lateral curvature, with an average penile curvature of 50° ± 28°. The average follow-up was 5.2 ± 1.8 months after the last administration of the drug. Ten percent of the patients exhibited improvement in the curvature, 60% had no change and 22% worsened; however, pain improved in 100% of the patients. The total average curvature was reduced to 47° ± 35°, with no statistical significance. Shirazi and colleagues reported the results of a controlled clinical trial, in which they could not demonstrate any improvement in penile curvature, pain intensity, consistency of the fibrous plaque, or sexual function in the verapamil study group patients compared with the control group.[25]

Chung and colleagues induced Peyronie's plaque in 12 adult male rats using an established PD animal model.[26] At 4 weeks, the rats were divided into group 1 with 0.1 mg/0.1 mL intralesional verapamil injected every second day for 2 weeks, group 2 with 0.1 mL intralesional normal saline injection, and group 3, which served as a control. At weeks 6 and 8, penile pressure was measured and serial immunohistochemical staining of penile tissue sections was performed. Intralesional injection of verapamil and normal saline resulted in macroscopic and microscopic changes in penile curvature and Peyronie's plaque size. Decreased collagen and elastin fibers were observed, together with a significant reduction in smooth muscle α-actin ($P < .05$). Changes were greater in group 1 than they were in group 2 ($P < .05$). Intralesional verapamil injection was associated with greater recovery of electrostimulated penile pressure, which is a surrogate of erectile function, compared with the saline and control groups. This research paradigm provides the opportunity to examine whether saline hydrodistention alone plays a significant role in inducing tissue remodeling.

Soh and colleagues compared the application of nicardipine with that of placebo.[27] They reported a significant reduction in the score of the sexual function index, the size of the plaque, and penile curvature, without a numerically significant reduction in curvature.

Over more than 5 decades, intralesional treatments have shown little promise for the treatment of PD, and there is no gold standard local treatment for this disease.[28] According to the treatment guidelines for PD published in 2010 and 2012, the intralesional treatment of the illness receives a grade B recommendation for interferon and grade C and D recommendations for calcium channel blockers, intralesional steroids, collagenase and orgotein.[6] A one-time application of onabotulinumtoxinA, which is indicated in other fibrotic pathologies, has been shown to cause an improvement in the signs and symptoms of patients with exaggerated scarring processes, without changing the tissue from a histological point of view. The average effective time is ≤ 6 months.[29]

PD shows great similarities regarding genetic expression with other processes in which fibrosis reduces the function of the affected organ. Dupuytren's disease shares a genetic overexpression of the transforming growth factor (TGF) β, which stimulates two types of receptors (1 and 2), and, with the contribution of the connective tissue and platelet-derived growth factors, favors the deposition of extracellular matrix and collagen while inhibiting the metalloproteinases that are in charge of breaking down collagen, to avoid excess scarring. This process allows the persistence of a dynamic form of over scarring and fibrosis in the palmar fascia and tunica albuginea.[30] Similar to PD and Dupuytren's disease, hypertrophic and keloid scars also involve the overexpression of TGF-β. The exposure of fibroblast cultures of these scars from animal and human models to the botulinum toxin reduces the expression of this facilitating fibrosis factor.[30]

OnabotulinumtoxinA derived from a gram-positive bacteria, Clostridium Botulinum with variety of usages in medicine for more than 20 years, demonstrating good efficacy and safety in various disorders. Many studies demonstrated that OnabotulinumtoxinA can inhibit the growth of hypertrophic scars, but the molecular mechanism for this action is unclear at all. Growing evidence suggests that onabotulinumtoxinA, influences cell apoptosis and proliferation and therefore may play a role in the expression of genes relevant to abnormal fibroblast proliferation.[29] TGF-β1 is known to be the most potent growth factor involved in wound healing and is believed to be the key regulator in the pathogenesis of hypertrophic scars . It is associated with an excessive deposition of scar tissue and fibrosis and modulates the expression of matrix metalloproteinases. Xiao and colleagues demonstrated that blocking TGF-β1 may attenuate the fibroproliferative response of hypertrophic scars and make its manipulation an attractive therapeutic strategy.[5] Connective transforming growth factor (CTGF) works as an independent mediator in fibrosis and collagen deposition in scars. OnabotulinumtoxinA may inhibit the growth of fibroblasts, which in turn may lead to a decrease expression of TGF-β1 protein and CTGF protein expression, therefore getting improvement of wound healing process. These results could partly explain the molecular mechanism for onabotulinumtoxinA based therapies for hypertrophic scar. This findings led the hypothesis that onabotulinumtoxinA reduces the expression of CTGF protein in fibroblast derived from hypertrophic scars. This have a relationship with TGF-β1 that decreased its expression too in fibroblast proliferation, also they saw tension diminishing around lesions by denervation stimulation due to onabotulinumtoxinA, not affecting novo fibrosis, no histologic changes around lesions. Therefore getting improvement of wound healing

process.[3-6]

Sahinkanat and colleagues use onabotulinumtoxinA in urethral strictures in rats compared with control group, finding less inflammation and fibrosis associated with less fibroblasts and collagen in tissues where have been applied botulinum toxin A.[31] However, two recent publications do not support any beneficial effect in the treatment of human keloids. Gauglitz and colleagues administered onabotulinumtoxinA in 4 patients (doses varying from 70 to 140 speywood units per session) injected directly into their keloids every 2 months for up to 6 months. Differences in height and volume were evaluated clinically and measured with a 3-D optical profiling system. Keloid-derived fibroblasts were treated with different concentrations of onabotulinumtoxinA, and expression of collagen as COL1A1, COL1A2, COL3A1, TGF-β1, TGF-β2, TGFβ3, fibronectin-1, laminin β2, and a-SMA was determined by real-time quantitative polymerase chain reaction (qRT-PCR) method. Intralesional administration of onabotulinumtoxinA did not result in regression of keloid tissue. They found no differences in expression of endothelial cell markers (ECM) markers, collagen synthesis, or TGFβ could be observed after onabotulinumtoxinA treatment of keloid fibroblasts. In addition, cell proliferation and metabolism of keloid fibroblasts was not affected by onabotulinumtoxinA treatment.[7]

Haubner and colleagues[8] tested patient-specific keloid tissue in a cell culture model to assess the effects of onabotulinumtoxinA incubation on cell proliferation and expression of the following cytokines and growth factors: interleukin-6, vascular endothelial growth factor and TGFβ. They found no evidence that these parameters of human keloid tissue were affected by onabotulinumtoxinA incubation.

In contrast, Wang and colleagues[6] found that, the S100A4 gene was highly expressed in keloid fibroblasts. S100 proteins belong to the superfamily of EF-hand calcium-binding proteins and have multifunctional roles in various cellular processes, including cell growth and differentiation, cell cycle regulation, apoptosis, transcription, and cell surface receptor activities. OnabotulinumtoxinA treatment can significantly affect the pathogenesis of keloids, particularly the invasive growth of keloid fibroblast cells, by influencing the regulation of some genes involved in cell invasion. Through gene microarray and qRT-PCR, they confirmed modulation of expression in 5 genes (S100A4, TGF-β1, VEGF, MMP-1, and PDGFA) in keloid-causing fibroblast cells treated with onabotulinumtoxinA.

We designed this protocol taking into account these precedents, together with extensive experience in the use of onabotulinumtoxinA in the treatment of multiple urological problems and hypertrophic and keloid scars.[3-6,29]

CONCLUSION

We considered the fact that intralesional treatments should require several administrations. Our results are promising with only one administration of the toxin. Our results suggest that onabotulinumtoxinA may be a new intralesional alternative therapy for PD. However, these findings should be evaluated with further randomized clinical trials including an adequate number of patients with multiple intralesional administrations with longer follow-up period, to confirm the potential therapeutic efficacy of this neurotoxin.

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CONFLICT OF INTEREST

None declared.

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