TITLE:

Fractional Carbon Dioxide Laser Therapy of the Vagina for Treatment of Urogential Symptoms in Women

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Study Protocol and Statistical Analysis Plan

Fractional Carbon Dioxide Laser Therapy of the Vagina for Treatment of Urogenital Symptoms in Women

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Table of Contents

SECTION I: STUDY OVERVIEW

Background and Rationale

Approximately half of post-menopausal women report vulvovaginal symptoms, including dryness, burning, itching, dyspareunia, and irritation[1]. These symptoms result from decreased circulating estrogen levels after menopause, which affects the vagina, vulva, musculature of the pelvic floor, endopelvic fascia, urethra, and bladder trigone. Vasculature and elasticity of these tissues decline with menopause, however these changes may reverse with treatment to restore estrogen[2]. Among women with vulvovaginal symptoms, 40% also reported overall sexual dysfunction, including issues with desire, arousal and orgasm[3].

As estrogen plays an important role in the function of the lower urinary tract throughout the premenopausal period, estrogen deficiency after menopause may predispose to lower urinary tract symptoms including urinary urgency, urge incontinence and stress incontinence[4]. Additionally, the incidence of urinary tract infection (UTI) rises dramatically in elderly women. Studies have shown that 15% to 20% of women aged 65 to 70 years and 20% to 50% of women over the age of 80 have bacteriuria[5]. Unlike the vasomotor symptoms of menopause, these vulvovaginal and urologic symptoms rarely self-resolve and often progress if untreated[6].

Currently available treatment options for these genitourinary symptoms of menopause (GSM) include use of lubricants with sexual intercourse, vaginal moisturizer a few times a week, twice weekly vaginal estrogen, a daily oral selective estrogen receptor modulator, or most recently vaginal dihydroepiandrosterone sulfate (DHEAS). All of these options require regular use of a medication, as a result several studies show high discontinuation rates of topical products[7].

More recently, fractional carbon dioxide (fCO_2) laser treatment of the vaginal tissue has been proposed as a treatment for genitourinary symptoms of menopause. Laser treatment is a longstanding option for skin discoloration and scarring, and has been shown to induce collagen production and tissue remodeling[8]. Use of laser in gynecology presumes that vulvovaginal symptoms are due to the thinning of the vaginal mucosa and loss of underlying collagen that occurs with menopause. Examination of pre- and post-treatment vaginal biopsies shows a significant increase in vaginal mucosal thickness without damage to surrounding tissue[9, 10]. Vaginal samples show an increase in lactobacillus colonization and decrease in pH[11]. Case report and case series data suggest that the treatment is well tolerated by patients and is not associated with serious adverse events[12]. Additionally, several case series demonstrate improvements in vulvovaginal[13, 14] and urinary symptoms[15], and in sexual function[16].

However, these studies are from a select few centers, are small, and are also biased in that the laser treatment is only available to patients who are able to pay for it, although patients have reported that they found the cost worthwhile[17]. In addition, most studies report only on vulvovaginal symptoms, despite increasing use of laser for urinary symptoms. Several recent editorials have called for larger studies with longer follow-up and high quality outcome measures[18-20].

At Beaumont's Women's Urology and Pelvic Health Center, we have the unique opportunity to provide fractionated carbon dioxide laser vulvovaginal treatment for a wide range patients with vulvovaginal, sexual and urologic complaints.

Objectives and Outcomes
This study will evaluate the effect of fractional carbon dioxide laser therapy of the vagina in female patients with urogenital symptoms.

Urogenital symptoms may include vaginal itching, burning, dryness, dyspareunia (difficult or painful sexual intercourse), dysuria (painful or difficult urination), nocturia (waking at night to urinate), incontinence (the involuntary loss of urine), urine frequency, urine urgency, and UTI.

Primary Objective	Evaluation of the subjective effects of fCO_2 laser therapy on overall urogentital symptoms.
Secondary Objective	Assess vaginal pH and bacteria, overall vaginal health, urinary symptoms, sexual function, UTI frequency, and mental health status.
Primary Outcome	The mean and median response to treatment as measured by the Patient Global Impression of Improvement (PGI-I) questionnaire
Secondary Outcomes	Change in vaginal pH and overall vaginal health on physical examination as measured by the Vaginal Health Inventory Score (VHIS)
	Change in urinary symptoms as measured by the Questionnaire for Urinary Incontinence Diagnosis (QUID), OAB symptoms as measured by the Overactive Bladder Questionnaire-Short Form (OAB-q SF), cough stress test and voiding diaries
	Change in sexual function, including dyspareunia symptoms, as measured by the Female Sexual Function Inventory (FSFI)
	Change in the frequency of UTI occurences
	Change in mental health status as measured by the Generalized Anxiety Disorder Questionnaire (GAD-7) and Patient Health Questionnaire (PHQ-8)
	Change in life and sexual impact of vulvovaginal skin symptoms as measured by the Incontinence Quality of Life Questionnaire (I-QOL)
	Change in the presence of lactobacillus, assessed by vaginal swab
Primary Endpoint	Week 16 (4 weeks post-treatment).
Secondary Endpoints	Primary and secondary outcomes will be evaluated again at Visit 4 (6-month follow-up)
	Primary and secondary outcomes will be evaluated again at Visit 10 (36-month follow-up)

SECTION II: METHODOLOGY

This is a prospective observational study of patients undergoing vaginal treatment with the fCO_2 laser for various urogenital symptoms. We anticipate enrolling approximately 100 women in this study. The majority of patients are expected to be recruited from the WUC and private Urology offices. Informed consent will be obtained before any study activities are conducted. Women who meet all of the eligibility criteria will be enrolled in the study. Three treatments with the fCO_2 laser to the vagina will be performed, approximately six weeks apart. The patient will follow up every three months for the first year, then every six months for the following two years, for a total of three years of follow-up.

At each follow-up visit, the patients will complete specified questionnaires. The patient will be offered a repeat treatment at 1, 2 and 3 years if initial treatment efficacy has decreased. A pelvic examination will be performed at Visits 4 (6-month follow up), 6 (12-month follow up), 8 (24-month follow up), and 10 (36-month follow up) and the clinician will complete a cough stress test and the VHIS, including vaginal pH.

Patients will be advised to refrain from:

- vaginal intercourse for 24 hours prior to all visits.
- starting vaginal or systemic hormone therapy or use of new vaginal lubricants during the first six months of the study.
- starting any other preventative therapies for UTIs including antiobiotic supression, cranberry supplements, or d-mannose during the first six months of the study.
- starting any new treatments (i.e. pharmacologic, physical therapy) for overactive bladder (OAB) or stress urinary incontinence (SUI) during the first six months of the study.

Study patients will not be compensated for their participation in the trial. However, all research related activities, specified on the table of events, will be provided at no cost to the study patient.

Inclusion/Exclusion Criteria

Inclusion criteria:

- Female
- ≥18 years and ≤90 years of age
- One or more of the following indications for fCO_2 laser treatment:
    - Genitourinary symptoms of menopause, including after natural, medically-induced, or surgical menopause
    - Vaginal dryness, burning, itching, or dyspareunia not related to menopause
    - Recurrent UTIs, defined as 4 or more in a year
    - Overactive bladder
- Must sign the informed consent form
- Must be willing to comply with the study protocol

Exclusion criteria:

- Contraindications to fCO_2 laser treatment, such as:
    - currently implanted synthetic pelvic mesh, sling, or tape
    - current or previous genital cancers
    - radiation to the vaginal or colo-rectal tissue
    - currently pregnant or less than 3 months following pregnancy
    - undiagnosed vaginal or cervical lesions

- Patients who have received vaginal fCO_2 laser treatment within the past 12 months
- Patients treated with vaginal estrogen within the past 3 months
- Patients with undiagnosed vaginal bleeding
- Active vulvar or vaginal infection, including herpes, candidiasis, etc.
- Current UTI, confirmed by positive urine culture and patient-reported UTI symptoms
- Pelvic or vaginal surgery within the past 9 months
- Pelvic organ prolapse beyond the introitus
- Patient possesses any other characteristics that, per the investigator's judgment, deems them unsuitable (i.e. may increase patient's risk, may affect the conduct of the study, etc.) for the treatment and/or study
- Participation in an investigational trial that used a study treatment, medication and/or biologic within 6 months or less prior to the date of the screening visit

Deferral

Patients may be deferred and rescreened at a later date under the following conditions:

- Patients with active vulvar or vaginal infections may be rescreened after treatment
- Patients with an active UTI may be rescreened after treatment
- Patients with undiagnosed vaginal bleeding may be rescreened after complete work-up, treatment and resolution of vaginal bleeding
- Patients pregnant within the past 3 months
- Patients who have received vaginal fCO_2 laser treatment within the past 12 months
- Patients treated with vaginal estrogen within the past 3 months
- Patients with recent pelvic or vaginal surgery within the past 9 months

Study Visits

Visit 0 – Baseline

Consent to send out a medical history form may be done by telephone script with all elements of information sheet included. Participant agreement is consent to send the medical history form. Once the participant is present for Visit 0, a formal consent will be performed for study participation. Informed consent will be obtained from all participants prior to conducting any study activities. Screening activities will be conducted to determine eligibility. Information will be provided from available medical records and patient self-report. Medical history, demographics, and current medications will be reviewed and recorded. Urine dipstick will be done to rule out active UTI. If UTI symptoms are reported and urine dipstick is positive for nitrates and/or leukocytes, a urine C&S will be sent to the laboratory. Urine pregnancy test will be done in women of childbearing capability. The patient will complete a three-day voiding within 1 week before Visit 1, which will be collected at Visit 1.

Visit 1 – Treatment 1

Visit 1 will, ideally, take place within 4 weeks of Visit 0. If Visit 1 occurs more than 4 weeks after Visit 0, then staff will review medical history and concomitant medications with the subject to ensure they are still eligible to participate in the study. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed (presence or absence of dysuria, foul change in urine odor, change in sediment, urgency, incontinence episodes). If a UTI is suspected, treatment will be deferred until the UTI is diagnosed and, if required, treated. Urine pregnancy test will be done in women of childbearing capability. Patient will complete the Vulvovaginal Symptom Questionnaire (VSQ), Incontinence Quality of Life (I-QOL) questionnaire, Questionnaire for female Urinary Incontinence Diagnosis (QUID), OAB-q SF, FSFI, GAD-7, and PHQ-8 prior to treatment. A vaginal exam, including a cough stress

test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus.The fCO_2 laser treatment will be performed, and the patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the VAS. Adverse events (AEs) will be assesed throughout the visit. The patient will complete a three-day voiding within 1 week prior to Visit 2, which will be collected at Visit 2.

Visit 2 – Treatment 2 (6 weeks +/- 2 weeks after Visit 1)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. Urine pregnancy test will be done in women of childbearing capability.The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus. The fCO_2 laser treatment will be performed, and patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the VAS. The patient will complete a three-day voiding diary within 1 week prior to Visit 3, which will be collected at Visit 3.

Visit 3 – Treatment 3 (6 weeks +/- 2 weeks after Visit 2)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. Urine pregnancy test will be done in women of childbearing capability.The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus. The fCO_2 laser treatment will be performed, and patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the VAS. The patient will complete a three-day voiding diary within 1 week prior to Visit 4, which will be collected at Visit 4.

In the event that scheduling difficulties result in treatment visits out of window, the treatment visits will take place no less than 4 weeks apart.

Phone Call (4 weeks +/- 1 weekafter Visit 3)
Research staff will contact the patient by phone to assess for AEs and administer the PGI-I for overall urogenital symptoms and Perception of Monetary Value.

Visit 4 - 6 Month Follow up (3months +/- 2 weeks after Visit 3)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus.

Visit 5 – 9 Month Follow up (3 months +/- 2 weeks after Visit 4)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. The patient will complete a three-day voiding diary within 1 week prior to Visit 6, which will be collected at Visit 6.

Visit 6 – 1 Year Follow up (3 months +/- 2 weeks after Visit 5)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. Urine pregnancy test will be done in women of childbearing capability.The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7, PHQ-8, and Perception of Monetary Value. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus. If the patient feels her symptoms have returned and she requests a repeat treatment this will be discussed with the investigator. If the investigator determines, in his/her clinical judgment, a treatment is appropriate the fCO_2 laser treatment will be performed.The patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the VAS.

Visit 7 – 18 Month Follow up(6 months +/- 2 weeks after Visit 6)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. The patient will complete a three-day voiding diary within 1 week prior to Visit 8, which will be collected at Visit 8.

Visit 8 – 2 Year Follow up (6 months +/- 2 weeks after Visit 7)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. Urine pregnancy test will be done in women of childbearing capability.The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus. If the patient feels her symptoms have returned and she requests a repeat treatment this will be discussed with the investigator. If the investigator determines, in his/her clinical judgment, a treatment is appropriate the fCO_2 laser treatment will be performed. The patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the VAS.

Visit 9 – 30 Month Follow up (6 months +/- 2 weeks after Visit 8)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assessed. The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. The patient will complete a three-day voiding diary within 1 week prior to Visit 10, which will be collected at Visit 10.

Visit 10 – 3 Year Follow up (6 months +/- 2 weeks after Visit 9)
Current medications and AEs will be assessed. Urine dipstick, and urine culture will be performed, if indicated. UTI symptoms will be assesses. Urine pregnancy test will be done in women of childbearing capability.The patient will complete a PGI-I for symptom improvement, VSQ, I-QOL, QUID, OAB-q SF, FSFI, GAD-7 and PHQ-8. A vaginal exam, including a cough stress test and VHIS, will be performed. A vaginal swab will be collected for wet mount and examined microscopically for presence of lactobacillus. If the patient feels her symptoms have returned and she requests a repeat treatment this will be discussed with the investigator. If the investigator determines, in his/her clinical judgment, a treatment is appropriate the fCO_2 laser treatment will be performed. The patient will score discomfort using the VAS. The provider will score the difficulty of administering the treatment using the

VAS. If a treatment is given, the participant will be instructed to follow up with the practitioner per standard of care. Visit 10 will be the final visit.

While in-person visits are preferrable, if the patient is unwilling or unable to attend a study visit (specifically visits 4-10) they will not be withdrawn from the study. In order to assess patient safety and provide continuing follow-up, the patient may be contacted by phone and/or mail. If necessary study information may be collected by either or both of these methods; phone (current medications, AE assessment) and/or mail (questionnaires, voiding diary).

Please see the next page for a schedule of visit activities.

Fractional Carbon Dioxide Laser Therapy of the Vagina for Treatment of Urogenital Symptoms in Women

Schedule of Activities

Visit:	Visit 0 Baseline	Visit 1 Tx 1	Visit 2 Tx 2	Visit 3 Tx 3	Phone call	Visit 4 6 Month Follow up	Visit 5 9 Month Follow up	Visit 6 1 Year Follow up	Visit 7 18 Month Follow up	Visit 8 2 Year Follow up	Visit 9 30 Month Follow up	Visit 10 3 Year Follow up
Visit Window:			*6 weeks±2 weeks from Visit 1*	*6 weeks±2 weeks from Visit 2*	*4 weeks±5 days from Visit 3*	*3 months±2 weeksfrom Visit 3*	*3 Months±2 weeks from Visit 4*	*3 Months±2 weeks from Visit 5*	*3 Months±2 weeks from Visit 6*	*3 Months±2 weeks from Visit 7*	*3 Months±2 weeks from Visit 8*	*3 Months±2 weeks from Visit 9*
Consent	X											
Inclusion/Exclusion*	X											
Medical History and Demographics*	X											
UTI Symptom Assessment	X	X	X	X		X	X	X	X	X	X	X
Urine Dipstick and Culture[a]	X	X	X	X		X	X	X	X	X	X	X
Urine Pregnancy Test[b]	X	X	X	X		X	X	X	X	X	X	X
Questionnaires[c]:												
FSFI		X	X	X		X	X	X	X	X	X	X
GAD-7		X	X	X		X	X	X	X	X	X	X
I-QOL		X	X	X		X	X	X	X	X	X	X
OAB-q SF		X	X	X		X	X	X	X	X	X	X
PGI-I			X	X	X	X	X	X	X	X	X	X
PHQ-8		X	X	X		X	X	X	X	X	X	X
Perception of Monetary Value					X			X				
QUID		X	X	X		X	X	X	X	X	X	X
VAS for Pain/Discomfort		X	X	X				X[d]		X[d]		X[d]
VSQ		X	X	X		X	X	X	X	X	X	X
VHIS[c], Vaginal Wet Mount, and cough stress test		X	X	X		X		X		X		X
Dispense Voiding Diary	X	X	X	X			X		X		X	
Collect/ Review Voiding Diary		X	X	X		X		X		X		X
Laser Treatment		X	X	X				X[d]		X[d]		X[d]
VAS for Procedure Difficulty[c]		X	X	X				X[d]		X[d]		X[d]
Assess AEs		X	X	X	X	X	X	X	X	X	X	X
Review Concomitant Medications*	X	X	X	X	X	X	X	X	X	X	X	X

[a]Culture will only be performed if dipstick is positive for nitrates and/or leukocytes

[b]Only in women of childbearing capability

[c]For full questionnaire/exam titles and descriptions please see reference the Measures section

[d]Only If patient wants laser treatment and investigator determines that laser treatment is appropriate

*In order to ensure eligibility, these items will be reassessed with extra precaution at Visit 1 if Visit 1 takes place > 4 weeks after visit 0

TREATMENT

Laser treatment will be performed as per routine protocol. This includes intra-vaginal treatment based on atrophy level as based on VHIS score and treatment number.

Treatment	Atrophy	Treatment 1	Treatment 2	Treatment 3
Intravaginal	Severe (VHIS 6-15)	Energy: 7.5-10 mJ Density: 10%	Energy: 10 mJ Density: 10%	Energy: 10 mJ Density: 10%
	Moderate (VHIS 16-20)	Energy: 10 mJ Density: 10%	Energy: 10-12.5 mJ Density: 10-15%	Energy: 10-12.5 mJ Density: 10-15%
	Non-atrophic (VHIS >20)	Energy: 10 mJ Density: 10%	Energy: 12.5 mJ Density: 10-15%	Energy: 12.5 mJ Density: 15%

Laser is delivered at 6 points at each level of the vaginal wall: 12, 2, 4, 6, 8, and 10, o'clock of Hart's line. Delivery begins at the most proximal and the wand is retracted by 1 cm and another row of laser treatment is delivered. Number of levels is determined by patient vaginal length.

MEASURES

Cough Stress Test

During a vaginal exam, while the patient is in lithotomy or frog-legged position, the provider will ask the patient to cough and will look for visual signs of a urine leak. Any urine loss observed is indicative of a positive cough stress test.

Female Sexual Function Index (FSFI)

This validated questionnaire is a brief multidimensional scale for assessing sexual function in women. The scale has received initial psychometric evaluation, including studies of reliability, convergent validity, and discriminant validity[24].

Generalized Anxiety Disorder 7-item (GAD-7)

This validated questionnaire is used for screening for anxiety with good reliability with sensitivity of 89% and specificty of 82%, for use in clinical practice and in research[25].

Incontinence-Quality of Life (I-QOL)

This validated questionnaire is a commonly used self-report quality of life measure specific to urinary incontinence. This tool can be used to assess the impact of urinary problems and their treatment. The I-QOL has 22 questions with the following 3 subscales: avoid and limiting behaviors (items), psychosocial impacts (9 items), and social embarassment (5 items). It is easily self-administered and takes about 5 minutes to complete.[23]

Overactive Bladder Questionnaire-Short Form (OAB-q SF)

This validated measure evaluates overactive bladder symptom severity and health related quality of life (Coyne, 2015). Statistically significant correlations have been found between the OAB-q SF scales and other patient reported outcomes instruments. For example, higher levels of depressive symptoms as measured by the Center for Epidemiologic Studies Depression Scale (CES-D) were linearly related to higher levels of Symptom Bother (r 0.31, $P<0.0001$) and lower levels of health related quality of life (HRQL) on the OAB-q SF (r 0.35, $P<0.0001$). Both the Symptom Bother and HRQL scales easily differentiated between controls without overactive bladder (OAB), continent patients with OAB and incontinent OAB patients ($P<0.0001$).

Patient Global Impression of Improvement (PGI-I)
The Patient Global Impression of Improvement (PGI-I) will be used to evaluate the patients' perceptions of overall improvement after treatment. The questionnaire has been validated in several urogenital conditions[21-23].

Personal Health Questionnaire Depression score (PHQ-8)
This validated questionnaire is used for monitoring depression severity and response to treatment and can be used to screen for depression in at-risk populations.

Perception of Monetary Value
The laser treatments will be provided at no cost to the study participants. This questionnaire will be completed at the primary endpoint (16 weeks) and the 12 month visit.

Questionnaire for Urinary Incontinence Diagnosis (QUID)
This validated measure is used to evaluate improvement in stress-predominant urinary incontinence in women undergoing non-surgical treatment. It has been found to be valid and responsive with internal consistency and acceptable psychometrics characteristics for use in clinical trials[24].

Vaginal Health Index Score (VHIS)
The VHIS is a tool, used by providers, to determine if vaginal atrophy is present. Providers perform a vaginal exam and give a score of 1-5 (abnormal to normal, respectively) on each of the following vaginal parameters: elasticity, secretion/fluid volume, vaginal pH, integrity of the epithelium, and lubrication/moisture of the vaginal wall. A VHIS score less than 15 is indicative of vaginal atrophy.

Visual Analog Scale (VAS) for Pain/Discomfort
Patients will record their average pain score during treatment from 0 (no pain) to 10 (most severe pain imaginable) on a Visual Analog Scale for pelvic pain and discomfort.

Visual Analog Scale (VAS) for Procedure Difficulty
Providers will record the difficulty of performing the treatment on a Visual Analog Scale from 0 (not at all difficult) to 10 (most difficult imaginable).

3-Day Voiding Diary
Patients will record each void and urine leak over a 3 day period. Patients will determine if each leak is a stress leak or an urge leak. They will score the level of urgency they experience with each void, based on the Indevus Urgency Severity Scale (IUSS).

Vulvovaginal Symptom Questionnaire (VSQ)
The VSQ was developed to determine the symptoms, emotions, life impact, and sexual impact of vulvovaginal symptoms on postmenopausal women. The questionnaire is comprised of twenty-one questions with "yes" or "no" responses.

SECTION III: POSSIBLE RISKS AND BENEFITS

Ethical Principles

This project will be approved by William Beaumont Hospital's Institutional Review Board (IRB). Adult patients will provide informed consent prior to any study activity. This study will be conducted pursuant to the Declaration of Helsinki: Recommendation Guiding the Medical Doctors in Biochemical Research Involving Human Subjects. Investigators will follow this clinical investigation plan at all times and follow requirements set by Federal Law and Institutional Review Board regulations at all times.

Risks of Study Procedures

Fractional Carbon Dioxide Laser Treatment

Risks of fractional carbon dioxide laser treatment to the vagina are minimal with no previous adverse events reported. Theoretically, risks may include vaginal irritation, bleeding, infection, and damage to surrounding tissue or structures. There also may be a burning sensation and itching, both mild in nature and resolving within 1-2 days.

Risk of Breach of Privacy and Confidentiality

There is a rare risk of breach of privacy and data confidentiality (release of information which personally identifies the patient). Confidentiality procedures will be strictly adhered to when transferring, managing, and analyzing study data. The patient will be assigned a unique study identification number and research information will be stored in a locked, secure cabinet in the Urology Resarch suite with access limited to authorized research personnel. Study data will be maintained in a password protected file on a shared network that can only be accessed by Urology research personnel.

Benefits

Patients may experience an improvement in urogenital symptoms. They will receive close clinical follow-up and monitoring throughout the study period.

SECTION IV: DATA SAFETY MONITORING PLAN

Safety Monitoring

Since the risks associated with this observational study are minimal and not outside the scope of routine care for adults with urological symptoms, oversight of data and safety will be primarily the responsibility of the PI and IRB in accordance with federal regulations. Adverse events and concomitant medications will be queried at each patient visit and reported to the PI in real time.

Data Management

Identifiers: Each enrolled patient will be assigned a unique study identification number (SIN). Protected Health Information (PHI) such as name, date of birth, or medical record number will not be used as the SIN. The SIN will consist of three digits, which will be assigned sequentially beginning with 001. A study enrollment log will be maintained and updated by study staff as patients are enrolled in the study. The enrollment log may contain the SIN and patient name, as well as additional identifiers.

Confidentiality: The enrollment log will be stored in a secure locked file cabinet and/or electronically on a drive with restricted access to Urology Research staff only. PHI that may be collected include patient name, address, telephone numbers, email address, medical record number and dates. The patient's birthdate, admission, discharge and procedures dates may also be collected. Only PHI essential to the research study will be utilized. All data collected during the course of the study will be stored in a secure, locked cabinet in the Urology Research

suite. To maintain confidentiality, only personnel directly involved in the research and the individuals and/or organization listed in the Consent Form and Authorization for Disclosure of Protected Health Information (also known as the Informed Consent Form) may have access to the research records. Medical and billing records collected for the purposes of the study will remain confidential, but may be disclosed (released) or used by the following representatives, specified in the Informed Consent Form:

- The investigators (study doctor/clinician, research staff)
- Beaumont Health System
- The patient's health insurance company and/or group health plans and their intermediaries (companies contracted to process claims)

If the patient requests information be shared with additional parties not listed on the Informed Consent (i.e. a primary care physician), a fully completed and signed Authorization for Disclosure of Subject Medical Information will be required prior to the release of study data.

Study data will be entered into a password protected database maintained on a local network within the Department of Urology. Access to the local network is restricted to Urology personnel, and access to the study database will be restricted to the study's key personnel only. Data will be entered by patient study identification number; names will not be linked with patient data in the database.

Our research practices safeguard the identity of study patients throughout the course of the study, including the dissemination of study findings. Study patients are not identified in any publication or other release of study results, data, and other information (such as professional writings, at professional meetings, in the sponsor's product information, and/or advertising or other promotional materials).

To assure patient privacy during the informed consent process, discussions may be conducted in a designated consenting room within the WUC or Urology Research Clinical suite. The rooms are separated from the reception, exam, lab, and office areas, and the doors are closed during discussions with research patients.

Data capture, verification, and disposition: Data may be captured from a number of sources including patient report, the electronic medical record, patient-administered questionnaires, clinical assessments, and diagnostic evaluations. All data entered on the case report form (CRF) is substantiated and verified by appropriate source documentation. Study data will be transcribed by study personnel from the source documents onto the CRF. Data recorded on the CRF will be entered into the database.

Clinical Monitoring Plan: Urology research nurse clinician(s) not participating in patient recruitment, enrollment, study visits or other study activities will be assigned to review completed case report forms and/or electronic data entry, data base review, IRB decisions, and investigator and clinical site records throughout the study. Additionally, patient charts, and clinical records will be reviewed to assure protocol adherence. Source documentation may also be verified.

He/she will review and verify the accuracy of data collection and documentation at the following time points:

- After the first patient has completed Visit 3
- After the first 6 patients have completed Visit 3
- After 20 patients have completed the 16-week phone call visit
- After 40 patients have completed the 16-week phone visit
- After 60 patients have completed the 16-week phone visit
- After 80 patients have completed the 16-week phone visit
- At the end of the study
- Additional monitoring may occur as needed

Patient records will be reviewed to verify CRF completion and delinquencies, and capture protocol deviations and frequency of unanticipated problems/adverse events. In order to identify, evaluate, and prevent adverse events, the total number of events will be reviewed, as well as the details of each event, including visit number and severity. Data will be reviewed to identify trends and possible concerns. Enrollment data will direct recruitment efforts and assist in study planning.

The overall objectives of routine monitoring are to:

- Document clinical study progress.
- Document that the protocol and associated forms are current.
- Obtain and review current clinical data, reports, and source documents.
- Review the overall study status including verification of the study files. All required documents and records should be accurate, complete, and current.
- Confirm that all patients have signed the informed consent form.
- Confirm that all enrolled patients have met the eligibility criteria.
- Verify accuracy of transferring data from source document(s) to the case report forms (CRFs) to the database.
- Confirm complete, accurate, and timely event reporting.
- Confirm adequacy of staff and facilities.
- Review communication records.
- Verify identification and reporting of protocol violations.

Outcomes of all scheduled and unscheduled study monitoring will be documented. Follow-up action items will be included in the report. The monitoring summary report will be forwarded to the PI and research coordinator.

Quality Control and Quality Assurance

The PI or designee has primary responsibility for QC/QA activities of the data. Beaumont's Research Institute also audits investigator-initiated studies to ensure compliance with consent processes and reporting requirements. The key QC/QA activities for the study will be:

- Clearly formatted and carefully constructed Case Report Forms (CRF);
- Sign-Off Procedures for all CRFs;
- Verification of patient eligibility at each visit

A database will be created utilizing RedCap, MS Excel, or equivalent software. Only designated study personnel will have access to the database, which will be stored on the Urology Research Department's shared drive. Access to the computerized system will be password protected. To ensure accuracy of data entry, database entries will be cross-checked against source

documents. The shared drive is backed up nightly to ensure minimal data loss, even in the most catastrophic system failure. The database will be active until all study visits are completed, final monitoring activities have been conducted, and all data queries have been resolved. After that time, the database will be locked, prohibiting any changes to the data.

All study records, including both paper and electronic, will be stored in accordance with all federal and institutional requirements, including, but not limited to, the HIPAA Privacy Rule, the Food and Drug Act, and Medicare policy. The stored data will be kept in a secure, protected manner. All records will be retained, at a minimum, for eleven years beyond study completion.

SECTION V: STATISTICAL CONSIDERATIONS AND DATA ANALYSIS

This is a novel observational study without a comparison group. Therefore, a power calculation is not necessary. Descriptive statistics will be given for all data collected. Patients will be analyzed and described overall as a group, and subgroup analyses will be performed by presenting complaint(s). Missing data will be considered missing and no interpolations or substitutions will be performed. Categorical characteristics will be summarized using frequencies and percentages. Continuous characteristics will be summarized using mean and standard deviation for normally distributed variables and median and 25th and 75th percentiles for those not normally distributed. The changes in the continuous measures from baseline (prior to device implant) to Visit 4 may be examined using paired t tests. We will explore relationships within this group of patients with possible complications using appropriate methods. SAS for Windows version 9.3 Cary, NC or equivalent will be used for all analyses.

SECTION VI: REFERENCES

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