European Review for Medical and Pharmacological Sciences 2024; 28: 814-821

Hybrid cooperative complexes to decrease VAS score and enhance sexual function in women with vulvar lichen sclerosus

M. TEDESCO[1], L. ALEI[1], A. BONADIES[1], T. PALLARA[1], P. PARISI[1], A. LATINI[2], B. BELLEI[3], F. SPERATI[4], E. MIGLIANO[1]

[1]Department of Plastic and Regenerative Surgery, San Gallicano Dermatological Institute IRCCS, Rome, Italy
[2]STI/HIV Unit, San Gallicano Dermatological Institute IRCCS, Rome, Italy
[3]Laboratory of Cutaneous Physiopathology and Integrated Center of Metabolomics Research, San Gallicano Dermatological Institute, IRCCS, Rome, Italy
[4]UOSD Clinical Trial Center, Biostatistical and Bioinformatic, San Gallicano Dermatological Institute IRCCS, Rome, Italy

Abstract. – OBJECTIVE: Lichen sclerosus is a chronic, inflammatory, progressive skin disease predominantly affecting anogenital areas. Vulvar lichen sclerosus (VLS) is one of the most common conditions treated in vulvar clinics; most patients report distressing symptoms of itching, burning, stinging, and pain (particularly during or after sexual intercourse). A preliminary, prospective, single-center study was performed to investigate the efficacy of hyaluronan hybrid cooperative complex (HCC) comprising high and low molecular weight hyaluronic acid to treat menopausal women with VLS.

PATIENTS AND METHODS: Patients (N = 30) received two HCC injections at 32 mg/ml (one month apart). At baseline and one and six months after treatment, patients completed validated psychometric questionnaires to assess their self-reported pain, itching, and dryness using the Visual Analogue Scale (VAS) and sexual function by the Female Sexual Function Index (FSFI).

RESULTS: After treatment with HCC, no side effects or complications were reported. VAS scores showed a trend towards reduced pain and itching intensity, and there was a statistically significant reduction in median VAS score for dryness at follow-up *vs.* baseline (p=0.038). For sexual function, there was a statistically significant improvement in lubrication (p=0.001) and orgasm (p=0.001) FSFI domains.

CONCLUSIONS: Overall, this preliminary study demonstrated the promising efficacy of HCC in menopausal women with VLS without side effects.

Key Words:
Vulvar lichen sclerosus, Menopausal women, Hyaluronan hybrid cooperative complex, VAS, FSFI, Rare dermatological disease, Regenerative therapy.

Introduction

Lichen sclerosus is a chronic, inflammatory, progressive skin disease mainly affecting anogenital areas[1-4]. Although vulvar lichen sclerosus (VLS) can arise at any age, it more commonly occurs in women (during prepuberty, perimenopause, or postmenopause) compared to men[5]. VLS is one of the most common conditions treated in vulvar clinics[1,6]. Estimates for the prevalence of VLS range from 1 in 300 to 1 in 1,000 patients referred to the hospital[5]. A Dutch pathology registry data[7] showed that the incidence rate of VLS increased from 7.4 per 100,000 woman-years in 1991 to 14.6 per 100,000 woman-years in 2011. VLS primary lesions appear as ivory or porcelain-white flat spots that can merge into pale, thin patches and plaques[1]. Some patients also have erythema, ecchymosis, and itching-related excoriations, and hyperkeratosis is prominent in a few patients[1]. VLS characteristically affects the inter-labial sulci, labia minora and majora, and clitoris and clitoral hood[1].

Most patients with VLS report distressing symptoms of itching, burning, stinging, and pain, particularly during or after sexual intercourse[1]. Other symptoms include chronic inflammation, anatomic changes, and the presence of erosions and fissures, which are the predominant cause of sexual pain and apareunia[1]. Despite physically and emotionally distressing symptoms, the clinical diagnosis of VLS is often delayed[1,5]. Furthermore, women with VLS have a lifetime risk of 2-5% of developing squamous cell carcinoma, and ≤65% of vulvar cancers arise in patients with a history of VLS[1].

Corresponding Author: Marinella Tedesco, MD; e-mail: marinella.tedesco@ifo.it

Although topical corticosteroids (e.g., clobetasol dipropionate 0.05% cream or preferably ointment) are standard of care and first-line treatment for VLS, prolonged use (>12 weeks) may result in dermal atrophy[1,2,5,6]. A safe and effective alternative to corticosteroids may be topical calcineurin inhibitors (e.g., tacrolimus or pimecrolimus)[5]. However, most second-line therapies require further investigation[6]. Surgery must be required to address some anatomical changes[5].

Autologous platelet-rich plasma (PRP) and autologous fat injection (lipofilling) were investigated in preliminary studies[7-10] as a treatment for tissue repair, including VLS regeneration, but is unsuitable for patients with platelet disorder, neoplastic disease, or those who are unable to take anticoagulants or antiplatelets[8]. Hyaluronic acid is a naturally occurring biodegradable polymer that can be used as a dermal filler[11]. In a phase 2 pilot study[12] of 20 postmenopausal women with vulvovaginal atrophy (VVA) and a history of breast cancer, treatment with autologous PRP combined with hyaluronic acid resulted in improved symptoms of vaginal dryness and dyspareunia. A novel formulation of HA composed of hybrid cooperative complexes (HCC) of low and high molecular weight HA and based on NAHYCO™ technology showed the ability to improve skin functions and remodeling. Studies[13] also supported the efficacy of HCC to stimulate elastin and collagen expression by keratinocytes and fibroblast, contributing to ameliorating skin extracellular matrix and global homeostasis. In a case series[14] of 26 women with VVA, treatment with HCC improved genital symptoms and sexual function. Furthermore, in a preliminary single-center study[8] of 20 women (aged 21-78 years) with VLS, HCC significantly reduced itching ($p \leq 0.001$), pain ($p=0.031$), and the sensation of burning ($p=0.004$) at six months *vs.* baseline.

A preliminary, prospective, single-center study was performed to investigate the efficacy of HCC comprising high and low molecular weight hyaluronic acid to treat menopausal women with VLS. Efficacy was determined by the impact of HCC treatment on self-reported pain, itching, dryness, and sexual function.

Patients and Methods

Ethics Approval

Approval for the treatment of VLS using Hybrid Cooperative Complexes (HCC) of hyaluronic acid (IBSA Farmaceutici Italia Srl, Lodi, Italy) was previously obtained from a local ethics committee (Protocol number, RS 1563/21). The single-center study was conducted at the Department of Plastic and Regenerative Surgery of San Gallicano Dermatological Institute IRCCS, Rome, Italy, between November 2021 and December 2022, and performed according to the Consolidated Standards of Reporting Trials guidelines and the Declaration of Helsinki. All patients involved provided informed consent to participate in this study and to use their data for scientific research.

Eligibility Criteria

Female patients eligible for the study had a histopathologic diagnosis of VLS and were otherwise in good health. Patients on steroid treatment were permitted to participate in the study but had to discontinue steroid use for the duration of the study. Patients who previously had regenerative therapy were only eligible to participate if they had completed this treatment ≥1 year before study enrollment. Exclusion criteria included patients with collagen vascular disease or genital infections. Patients with allergies and previous reactions to fillers were also excluded from the study.

Study Design

Following detailed explanations of the procedure and possible side effects or complications, enrolled patients provided informed consent to participate in the study. Patients received a highly purified, thermally stabilized, high- and low-molecular-weight HCC of hyaluronic acid of non-animal origin without any chemical crosslinking agents at a dose of 32 mg/ml (IBSA Farmaceutici Italia Srl, Lodi, Italy). Patients received HCC injections at two separate hospital visits, one month apart (Figure 1). An anesthetic ointment was applied to the genital region 30 minutes before injection. A single syringe containing 2 mL of HCC was used per treatment and injected into the genital region of the patient with a 29-gauge needle and an intradermal wheal technique (0.25 mL per injection site) distributed as follows: two into the posterior fourchette, two into the labia minora, two into the vulvar vestibule, and two near the clitoris.

Study Outcomes

At baseline (timepoint 0, T0), one month after treatment completion (T1), and six months after

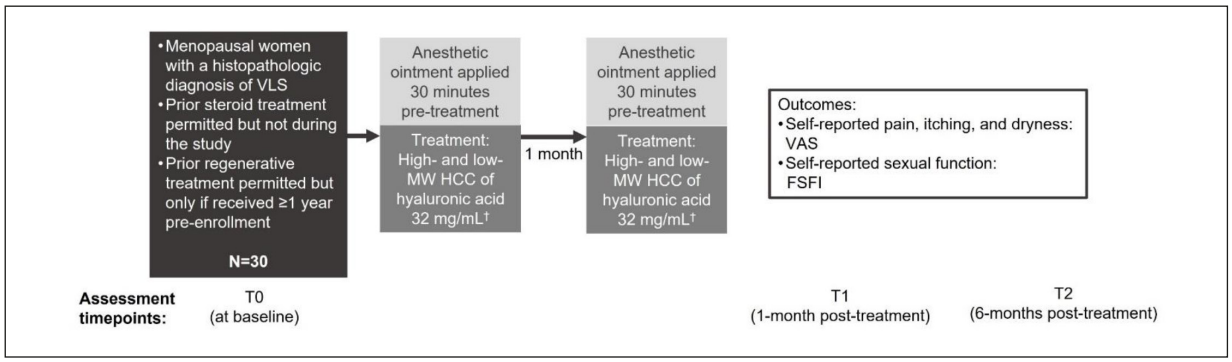

Figure 1. Study design. †A single syringe filled with 2 mL of HCC was used per treatment, and HCC was injected into the genital region of the patient with a 29-gauge needle and an intradermal wheal technique (0.25 mL per injection site) distributed as follows: 2 into the posterior fourchette, 2 into the labia minora, 2 into the vulvar vestibule, and 2 near the clitoris. FSFI, Female Sexual Function Index; HCC, Hybrid Cooperative Complexes; MW, molecular weight; N, number of patients; T, timepoint; VAS, Visual Analogue Scale; VLS, Vulvar lichen sclerosus.

treatment completion (T2), patients completed validated psychometric questionnaires to assess their self-reported pain, itching, dryness, and sexual function. Patients graded the intensity of their symptoms (pain, itching, and dryness) on a 0- to 10-point Visual Analogue scale (VAS) questionnaire, with 0 indicating no symptoms (no pain, itching, dryness) and 10 indicating maximum symptoms[15,16].

Patients also rated their sexual function at T0, T1, and T2 using the 19-item Female Sexual Function Index (FSFI) questionnaire[17] consisting of the following domains: desire, arousal, lubrication, orgasm, satisfaction, and pain. Each domain includes the following weight coefficients and number of items/questions: desire (2 items*0.6), arousal (4 items*0.3), lubrication (4 items*0.3), orgasm (3 items*0.4), satisfaction (3 items*0.4), and pain (3 items*0.4)[17]. A summary of the FSFI questionnaire with included items or questions is shown in **Supplementary Table I**.

Statistical Analysis

For categorical variables, absolute and relative frequencies were assessed. For continuous variables, however, means, standard deviations (SD), median values, and interquartile range were calculated. In addition, the Kolmogorov-Smirnov normality test was performed for all continuous variables. Variables (found not to have a normal distribution) were assessed using the non-parametric Friedman test. The Friedman test was used to evaluate differences in overall scores between study time points (T0, T1, T2), with statistically significant differences determined by p-values ≤ 0.05. All statistical analyses were performed using GraphPad Prism software version 9.5.1 (GraphPad Software, San Diego, CA, USA).

Results

Baseline Demographic and Clinical Characteristics

A total of 30 female patients were enrolled in the study. One patient subsequently withdrew from the study for personal reasons; therefore, 29 patients were included in the data analyses. The mean age of 29 patients was 62.4 (range: 54-79) years (Table I). At baseline (T0), all patients experienced dryness, and most patients also had itching (93%) and pain (86%). Most patients (72%) also reported being sexually active. No complications or side effects were reported in the study.

Self-Reported Pain, Itching, and Dryness

All VAS domain scores for pain, itching, and dryness, as well as total VAS score, showed a trend towards a reduction in symptom intensity at T1 (one month after treatment) and T2 (six months after treatment) *vs.* baseline (Table II). The reduction in symptom intensity according to the VAS score was statistically significant for dryness; the median (interquartile range, IQR) VAS score for dryness was significantly reduced from 7 (5, 8) at T0 to 6 (5, 8) at T1 and 5 (4, 8) at T2 (p=0.038; Table II).

Table I. Baseline demographic and clinical characteristics.

	Female patients N = 29
Age, years, mean (range)	62.4 (54-79)
Symptoms, n (%)	
Dryness	29 (100.0)
Itching	27 (93.1)
Pain	25 (86.2)
Sexually active, n (%)	
Yes	21 (72.4)
No	2 (6.9)
N/A	6 (20.7)
Highest education level, n (%)	
High school diploma	12 (41.4)
University graduate	10 (34.5)
Less than high school	5 (17.2)
N/A	2 (6.9)
Marital status, n (%)	
Married	20 (69.0)
Divorced	3 (10.3)
Cohabitee	1 (3.4)
Partner (not cohabitee)	1 (3.4)
Single	1 (3.4)
Widow	1 (3.4)
N/A	2 (6.9)
Occupation, n (%)	
Employee	7 (24.1)
Retired	7 (24.1)
Housewife	3 (10.3)
Manager	3 (10.3)
Self-employed	3 (10.3)
Artisan	1 (3.4)
Teacher	1 (3.4)
Other	2 (6.9)
N/A	2 (6.9)
Religious, n (%)	
Yes	8 (27.6)
No	14 (48.3)
N/A	7 (24.1)

n, number of patients; N, total number of patients; N/A, not available.

Table II. Statistical analysis of VAS domain scores (pain, itching, dryness) over time.

VAS score	Median (IQR)			Friedman test *p*-value
	T0	T0	T2	
Pain	5 (3-7)	3 (0-6)	3 (0-6)	0.121
Itching	5 (1-8)	3 (2-6)	3 (1-6)	0.076
Dryness	7 (5-8)	6 (5-8)	5 (4-8)	0.038*

A lower VAS score indicates lower symptom intensity. Descriptive statistics were used to establish median (IQR) values for all patients (N=29). One patient with only 1 VAS measurement at T0 (but not at T1 and T2) was excluded from Friedman's test as this statistical test cannot compute missing values. *The change in VAS scores for dryness over time was found to be statistically significant ($p < 0.05$). IQR, interquartile range; T, timepoint, VAS, Visual analogue scale.

Self-Reported Sexual Function

Most of the FSFI scores for the different domains of the FSFI questionnaire (desire, arousal, lubrication, orgasm, satisfaction, and pain) showed a trend toward a higher score at T1 and T2 (one month and six months after treatment), which shows that HCC treatment improved sexual function (Table III). More specifically, there was a significant improvement in the lubrication and orgasm domains of the FSFI; the median (IQR) FSFI score for both lubrication and orgasm improved from 0 (0, 3) at T0 to 2 (0, 4) at T1 and T2 (p=0.001 for both domains). For lubrication, the improvement in FSFI scores following treatment *vs*. baseline was statistically significant across all lubrication-related items or questions of the questionnaire (Table III) that covered lubrication frequency (p=0.013), lubrication difficulty (p=0.003), frequency in maintaining lubrication (p=0.001), and difficulty in maintaining lubrication (p=0.001). For orgasm, the improvement in FSFI scores was statistically significant for items or questions of the FSFI questionnaire covering difficulty and satisfaction of achieving an orgasm (p=0.030 and p=0.041, respectively) but not for orgasm frequency (p=0.307). At an individual FSFI item level, satisfaction with the amount of closeness with the patient's partner was also significantly improved (p=0.032). However, there was no change in the median FSFI score for satisfaction with sexual relationships (p=0.119) or overall sex life (p=0.241).

Although no statistically significant increase in the total FSFI score (p=0.756) was reached, for patients with a total FSFI score <20 at baseline

Table III. Statistical analysis of FSFI scores for each question in the FSFI questionnaire over time.

Domain	Item	Theme of question	Median (IQR)			Friedman test p-value
			T0	T1	T2	
Desire	Q1	Frequency	2 (1-3)	2 (1-3)	2 (1-3)	0.677
	Q2	Level	2 (1-3)	2 (1-3)	2 (1-3)	0.497
Arousal	Q3	Frequency	1 (0-4)	2 (0-4)	2 (0-4)	0.124
	Q4	Level	0 (0-3)	2 (0-3)	2 (0-3)	0.074
	Q5	Confidence	2 (0-3)	2 (0-3)	2 (0-3)	0.073
	Q6	Satisfaction	0 (0-3)	2 (0-4)	2 (0-4)	0.098
Lubrication	Q7	Frequency	0 (0-2)	1 (0-3)	1 (0-3)	**0.013***
	Q8	Difficulty	0 (0-3)	1 (0-3)	1 (0-3)	**0.003***
	Q9	Frequency in maintaining	0 (0-2)	1 (0-4)	1 (0-3)	**0.001***
	Q10	Difficulty in maintaining	0 (0-3)	2 (0-4)	2 (0-3)	**0.001***
Orgasm	Q11	Frequency	0 (0-4)	2 (0-4)	2 (0-4)	0.307
	Q12	Difficulty	0 (0-3)	2 (0-3)	3 (0-3)	**0.030***
	Q13	Satisfaction	0 (0-4)	2 (0-4)	3 (0-4)	**0.041***
Satisfaction	Q14	Amount of closeness with partner	0 (0-4)	2 (0-4)	3 (0-4)	**0.032***
	Q15	Sexual relationship	3 (1-4)	3 (2-4)	3 (2-4)	0.119
	Q16	Overall sex life	3 (1-4)	3 (2-4)	3 (2-4)	0.241
Pain	Q17	Frequency during vaginal penetration	0 (0-1)	0 (0-2)	0 (0-1)	0.261
	Q18	Frequency after vaginal penetration	0 (0-1)	0 (0-1)	0 (0-1)	0.050
	Q19	Frequency during/after vaginal penetration	0 (0-1)	1 (0-2)	0 (0-2)	0.109
Desire			2 (2-4)	2 (1-4)	2 (1-4)	0.911
Arousal			1 (0-5)	2 (0-4)	2 (0-4)	0.635
Lubrication			0 (0-3)	2 (0-4)	2 (0-4)	**0.001***
Orgasm			0 (0-3)	2 (0-4)	2 (0-4)	**0.001***
Satisfaction			2 (1-5)	4 (2-5)	4 (1-5)	0.175
Pain			0 (0,1)	0 (0-2)	0 (0-2)	0.391
Total score			5 (4-22)	10 (4-20)	13 (4-21)	0.756
Total score if FSFI score < 20 at T0			4 (3-5)	6 (4-14)	4 (4-14)	**0.042***
Total score if FSFI score = 20-30 at T0			23 (22-26)	20 (19-23)	23 (21-28)	**0.028***

A lower VAS score indicates lower symptom intensity. Descriptive statistics were used to establish median (IQR) values for all patients (N=29). One patient with only 1 VAS measurement at T0 (but not at T1 and T2) was excluded from Friedman's test as this statistical test cannot compute missing values. *The change in VAS scores for dryness over time was found to be statistically significant ($p < 0.05$). IQR, interquartile range; T, timepoint, VAS, Visual analogue scale.

(20 patients, but only 19 included in the Friedman's test as one patient had missing values at T1 and T2), there was a statistically significant difference in median total FSFI score between the three timepoints (p=0.042; Table III) with an increase in median FSFI score from 4 at T0 to 6 at T1 but no change at T2. For patients with total FSFI score 20-30 at baseline (9 patients), there was a statistically significant difference in median FSFI score between the three time points (p=0.028) with a decrease in median FSFI score from 23 at T0 to 20 at T1 but no change at T2.

Discussion

In this preliminary, prospective, single-center study, 29 women (54 to 79 years of age) with a histopathologic diagnosis of VLS who were otherwise in good health received two HCC injections comprising high and low molecular weight hyaluronic acid (1 month apart). Participants completed validated psychometric questionnaires to assess their self-reported pain, itching, dryness, and sexual function at T0 (baseline), T1 (one month after treatment completion), and T2 (six months after treatment completion). There was a statistically significant reduction in the median VAS score for dryness at T1 and T2 *vs.* baseline (p=0.038), but not for pain and itching, even though their median VAS scores showed a trend towards a reduction in symptom intensity. Regarding sexual function, there was only a statistically significant improvement in lubrication (p=0.001) and orgasm (p=0.001) domains of the FSFI, including all lubrication-related items or questions of the questionnaire and items of the FSFI questionnaire covering difficulty and satisfaction of achieving an orgasm, but not for orgasm frequency. There was an increase in the median total FSFI score from baseline to T1 and T2, but this was not statistically significant (p=0.756).

The outcomes from this preliminary study are promising and highlight the potential benefit of HCC in menopausal women with VLS. The results are comparable with our prior study[8] in which 20 females (age range, 21-78 years) with VLS received monthly HCC injections three times. In that study, however, symptoms of itching, pain, burning sensation, and dyspareunia were measured by the number of patients who answered 'yes' or 'no' to having those symptoms rather than a score of the severity of those symptoms; there was a statistically significant reduction in the number of patients who had itching ($p\leq$0.001), pain (p=0.031), and burning sensation (p=0.004) six months after completing treatment[8]. In addition, patients in that study who already had low FSFI scores (<20 or 20-26) at baseline, showed reduced FSFI scores (indicating worse sexual function) six months after completing treatment[8]. In contrast, those with FSFI score >26 at baseline showed a trend towards higher FSFI scores six months after completing treatment, although this was not statistically significant (p=0.084)[8]. Conversely, in our current study, the total FSFI score was statistically different for the three time points for patients with total scores <20 at baseline and patients, with total scores of 20-26 at baseline, but not for all patients (irrespective of total FSFI score at baseline). The median FSFI score at six months after treatment was the same as at baseline for patients with FSFI score <20 at baseline and patients with FSFI score of 20-26 at baseline. However, there was a trend towards an increase in median FSFI score at one month after treatment for patients in our study with a total FSFI score <20 at baseline and a trend towards a decrease in median FSFI score at one month after treatment for patients with total FSFI score of 20-30 at one month after treatment.

Menopausal women with VLS who got two HCC injections reported a promising improvement in dryness symptoms and an improvement in the lubrication and orgasm domains of sexual function without side effects. This backs up what was found in earlier studies of HCC injections in women with VLS[8] or vulvar vaginal atrophy, both of which have a wide age range[14]. Our study used a thermally stabilized HCC with low viscosity to enable optimal tissue diffusion and two different molecular weights[14]. Hyaluronan has previously been shown[18] to be useful for tissue hydration, activation of fibroblasts and keratinocytes, and to have antioxidant and anti-inflammatory effects. HCCs *vs.* linear hyaluronan and cross-linked hyaluronans were previously shown[18] to enhance the differentiation and proliferation of adipose-derived stem cells. Possible side effects of hyaluronans injections include erythema and short-term bruising[8,19].

Limitations

Dry needling has previously been shown[20] to improve localized musculoskeletal pain symptoms; this may have influenced patient outcomes in our study, but data on the maintenance of

response following dry needling are sparse. Furthermore, our study is limited due to its small sample size, the fact that it is a single-center study, and the lack of a comparative treatment arm to evaluate treatment response.

Conclusions

Despite study limitations, the promising efficacy of HCC comprising high and low molecular weight hyaluronic acid in menopausal women with VLS without side effects was demonstrated. At six months of follow-up, two monthly injections of HCC led to a statistically significant reduction in self-reported symptoms of dryness (as measured by the VAS) and a significant improvement in self-reported lubrication and orgasm aspects of sexual function (as measured by FSFI). Further work should include larger studies with a comparative treatment arm to confirm the efficacy and safety of HCC in women with VLS.

Conflict of Interest

A.L. received research funding from IBSA Farmaceutici Italia Srl.

Acknowledgements

The authors are grateful to Sally Hassan, Ph.D. of Papyrus Medical Communications Ltd, for helping in writing the first draft of the manuscript. Medical writing was sponsored by IBSA Farmaceutici Italia. The authors acknowledge Clara Cigni and Franco Grimolizzi (employees of IBSA Farmaceutici Italia Srl) for the revision and the scientific discussion.

Data Availability

The data that support study findings are available upon request from the corresponding author. These data are not publicly available due to privacy or ethical restrictions.

Funding

This study and manuscript were funded by IBSA Farmaceutici Italia Srl.

Ethics Approval

Approval for the treatment of VLS using Hybrid Cooperative Complexes (HCC) of hyaluronic acid (IBSA Farmaceutici Italia Srl) was previously obtained from a local ethics committee (Protocol number, RS 1563/21). The single-center study was conducted at the Department of Plastic and Regenerative Surgery of San Gallicano Dermatological Institute IRCCS, Rome, Italy, between November 2021 and December 2022 and performed according to the Consolidated Standards of Reporting Trials guidelines and the Declaration of Helsinki.

Informed Consent

All the patients involved provided informed consent to participate in this study and to use their data for scientific research.

ORCID ID

Marinella Tedesco: 0000-0001-5816-5834

References

1) Corazza M, Schettini N, Zedde P, Borghi A. Vulvar Lichen Sclerosus from Pathophysiology to Therapeutic Approaches: Evidence and Prospects. Biomedicines 2021; 9: 950-956.
2) Kirtschig G, Becker K, Gunthert A, Jasaitiene D, Cooper S, Chi C, Kreuter A, Rall K, Aberer W, Riechardt S, Casabona F, Powell J, Brackenbury F, Erdmann R, Lazzeri M, Barbagli G, Wojnarowska F. Evidence-based (S3) Guideline on (anogenital) Lichen sclerosus. J Eur Acad Dermatol Venereol 2015; 29: 1-43.
3) Fistarol SK, Itin PH. Diagnosis and treatment of lichen sclerosus: an update. Am J Clin Dermatol 2013; 14: 27-47.
4) Tedesco M, Garelli V, Elia F, Chicherchia G, Foddai ML, Latini A, Morrone A, Migliano E. Usefulness of video thermography in the evaluation of platelet-rich plasma effectiveness in vulvar lichen sclerosus: preliminary study. J Dermatolog Treat 2021; 32: 568-571.
5) Pérez-López FR, Ceausu I, Depypere H, Tamer Erel C, Lambrinoudaki I, Rees M, Schenck-Gustafsson K, Tremollieres F, van der Schouw Y T, Simoncini T. EMAS clinical guide: vulvar lichen sclerosus in peri and postmenopausal women. Maturitas 2013; 74: 279-282.
6) Singh N, Mishra N, Ghatage P. Treatment Options in Vulvar Lichen Sclerosus: A Scoping Review. Cureus 2021; 13: 13527-13533.
7) Bleeker MC, Visser PJ, Overbeek LI, van Beurden M, Berkhof J. Lichen Sclerosus: Incidence and Risk of Vulvar Squamous Cell Carcinoma. Cancer Epidemiol Biomarkers Prev 2016; 25: 1224-1230.
8) Tedesco M, Garelli V, Elia F, Sperati F, Biondi F, Mosiello L, Morrone A, Migliano E. Efficacy of injecting hybrid cooperative complexes of hyaluronic acid for the treatment of vulvar lichen sclerosus: A preliminary study. J Cosmet Dermatol 2023; 22: 449-457.
9) Tedesco M, Bellei B, Garelli V, Caputo S, Latini A, Giuliani M, Cota C, Chichierchia G, Romani C,

Foddai ML, Cristaudo A, Morrone A, Migliano E. Adipose tissue stromal vascular fraction and adipose tissue stromal vascular fraction plus platelet-rich plasma grafting: New regenerative perspectives in genital lichen sclerosus. Dermatol Ther 2020; 33: 14277-14281.

10) Lacci KM, Dardik A, Platelet-rich plasma: support for its use in wound healing. Yale J Biol Med 2010; 83: 1-9.
11) Fakhari A, Berkland C. Applications and emerging trends of hyaluronic acid in tissue engineering, as a dermal filler and in osteoarthritis treatment. Acta Biomater 2013; 9: 7081-7092.
12) Hersant B, SidAhmed-Mezi M, Belkacemi Y, Darmon F, Bastuji-Garin S, Werkoff G, Bosc R, Niddam J, Hermeziu O, La Padula S, Meningaud JP. Efficacy of injecting platelet concentrate combined with hyaluronic acid for the treatment of vulvovaginal atrophy in postmenopausal women with history of breast cancer: a phase 2 pilot study. Menopause 2018; 25: 1124-1130.
13) Stellavato A, La Noce M, Corsuto L, Pirozzi AVA, De Rosa M, Papaccio G, Schiraldi C, Tirino V. Hybrid Complexes of High and Low Molecular Weight Hyaluronans Highly Enhance HASCs Differentiation: Implication for Facial Bioremodelling. Cell Physiol Biochem 2017; 44: 1078-1092.
14) Garavaglia E, Sala C, Busato M, Bellia G, Tamburlin N, Massirone A. First Use of Thermal Stabilized Hyaluronic Acid Injection in One-Year Follow-Up Patients with Genitourinary Syndrome. Med Devices 2020; 13: 399-410.
15) Delgado DA, Lambert BS, Boutris N, McCulloch PC, Robbins AB, Moreno MR, Harris JD, Validation of Digital Visual Analog Scale Pain Scoring With a Traditional Paper-based Visual Analog Scale in Adults. J Am Acad Orthop Surg Glob Res Rev 2018; 2: 088-089.
16) Bizjak Ogrinc U, Sencar S, Luzar B, Lukanovic A. Efficacy of Non-ablative Laser Therapy for Lichen Sclerosus: A Randomized Controlled Trial. J Obstet Gynaecol Can 2019; 41: 1717-1725.
17) Rosen R, Brown C, Heiman J, Leiblum S, Meston C, Shabsigh R, Ferguson D, D'Agostino Jr R. The Female Sexual Function Index (FSFI): a multidimensional self-report instrument for the assessment of female sexual function. J Sex Marital Ther 2000; 26: 191-208.
18) Dicker KT, Gurski LA, Pradhan-Bhatt S, Witt RL, Farach-Carson MC, Jia X. Hyaluronan: a simple polysaccharide with diverse biological functions. Acta Biomater 2014; 10: 1558-1570.
19) Haneke E. Managing Complications of Fillers: Rare and Not-So-Rare. J Cutan Aesthet Surg 2015; 8: 198-210.
20) Leggit JC. Musculoskeletal Therapies: Acupuncture, Dry Needling, Cupping. FP Essent 2018; 470: 27-31.