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The influence of hormone replacement therapy on skin ageing
A pilot study

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Abstract

Objectives: We studied the effect of hormonal treatment on skin ageing in menopausal women. *Methods:* Twenty-four patients (45–68 years; mean age, 54.9 years) without hormone treatment for at least 6 months were included. Patients were assigned to three therapy groups: 1, oestrogen only (Estraderm TTS® 50) ($n = 6$); 2, transdermal oestrogen and progesterone (Estraderm TTS® 50 and 0.4 mg progesterone vaginal suppository) ($n = 7$); and 3, oral oestrogen and progesterone (2 mg Progynova® and 0.4 mg progesterone vaginal suppository) ($n = 8$). One group without therapy was included as a control group ($n = 3$). Treatment was continued for 6 months. Three patients, one from group 2 and two from group 3, discontinued therapy before the study endpoint. The following skin parameters were measured at monthly intervals during treatment: skin surface lipids, epidermal skin hydration, skin elasticity and skin thickness. Concomitant clinical evaluation included a subjective clinical evaluation form, a patient questionnaire and laboratory tests for oestradiol, progesterone and follicle stimulating hormone. *Results:* Mean levels of epidermal skin moisture, elasticity and skin thickness were improved at the end of treatment based on both subjective and objective evaluation in patients with hormone replacement therapy (HRT). Skin surface lipids were increased during combined HRT, which may reflect stimulatory effects of the progestagen component on sebaceous gland activity, while oestrogen alone has a sebum-suppressive action. In the HRT groups, the questionnaire for climacteric complaints demonstrated significant improvements, while laboratory tests showed increases in oestradiol and progesterone and decreases in FSH. *Conclusions:* HRT with the mentioned regimes significantly improved parameters of skin ageing.

Keywords: Hormone replacement therapy; Skin ageing; Non-invasive measurements

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1. Introduction

Like all other tissues, skin undergoes degenerative processes during ageing. Although skin ageing is associated with increased rates of skin disorders and skin tumours, and sometimes with psychological distress caused by a deterioration in appearance, the main focus of public medicine is not on skin ageing but on other age-associated chronic disorders such as arthritis, heart disease and cancer [1,2]. At present, most women in developed societies can expect to spend one-third or more of their lifetime in the postmenopausal period [2]. Skin ageing, therefore, becomes increasingly important; it is still a matter of debate, however, whether hormone replacement therapy (HRT) also improves the various symptoms of skin ageing in postmenopausal women.

The effects of HRT on the skin have not been studied in detail despite the importance of dermatological aspects of ageing in this population [3–6].

Cutaneous ageing is the result of a combination of chronological and environmental (e.g. nicotine, pollution, UV-induced ageing) factors, and hormonal ageing [7].

Skin is a target organ for various hormones. Hormonal action requires the binding of the hormone to specific receptors. Oestrogen receptors as well as other hormone receptors have been isolated and characterized in the human skin [8]. Oestrogen receptors are to be found in skin structures associated with skin ageing processes. A decrease in oestrogen influences might thus induce a reduction of those skin functions that are under oestrogen control.

In clinical terms, many females experience a sudden onset of skin ageing symptoms several months after menopause. One of the first symptoms of skin ageing women experience after menopause is an increase in skin dryness, followed by a decrease in skin firmness and elasticity. The increasing looseness of the skin outweighs other symptoms such as wrinkles at that stage of reduced hormonal action. These symptoms correspond to changes in collagenous and elastic fibres that have been reported to be due to oestrogen deficiency [9]. A significant decrease in skin collagen starting at the menopause has been demonstrated [1]. Among the various types of collagen, types I and III are of major relevance [10]. Both types of collagen change during the ageing process. Type I collagen represents the predominant collagen type in adult human skin, whereas type III collagen, also widely distributed throughout the body (being the second most frequent collagen in the adult human skin), predominates in tissues of foetuses. Oestrogens stimulate the synthesis, maturation and turnover of collagen, increase the synthesis of hyaluronic acid, and promote water retention [11].

In view of these findings, we performed the present ad hoc pilot study, comparing the effects of three different forms of HRT on various parameters of skin ageing.

2. Materials and methods

2.1. Patients

Twenty-four menopausal women aged 45–68 years (mean age, 54.9 years), who had received no hormonal treatment for a minimum of 6 months, who had been amenorrhoeic for at least half a year and whose bone density values were within the normal range, were included in the study. On average, the study participants were 8.2 years after their last menstrual period (0.5–30 years). Further menopausal criteria were their initial hormone levels: low oestradiol (E_2) (< 45 pg/ml) and high follicle stimulating hormone (FSH) (> 30 mU/ml) serum levels (Table 1). Of the 24 patients, eight had undergone a hysterectomy and three an oophorectomy. The probands were female volunteer outpatients who had been referred to our centre from the gynaecological department and who had all given their informed consent to the treatment and the investigations.

2.2. Study design and treatment

In this ad hoc study, the effects of three different HRT regimes on different parameters of skin ageing were compared over a period of 6 months. The patients were assigned to four groups: group

Table 1
Initial and final hormone levels[a]

		Group 1			Group 2			Group 3			Group 4		
		Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*
E_2 (pg/ml)	Mean	29.43	45.83	0.12	16.33	49.80	0.08	25.83	59.33	0.08	37.33	18.00	n.s.
	S.D.	13.29	23.04	n.s.	7.63	46.83	n.s.	21.21	34.10	n.s.	28.50	5.57	
FSH (mU/ml)	Mean	45.98	40.13	0.25	63.15	43.68	0.08	69.52	50.12	0.08	35.13	41.93	n.s.
	S.D.	15.42	14.03	n.s.	34.94	20.18	n.s.	15.31	26.41	n.s.	23.75	19.03	
Progesterone (ng/ml)	Mean	0.29	0.31	0.59	0.29	1.93	0.04	0.37	0.82	0.25	0.47	0.24	n.s.
	S.D.	0.22	0.29	n.s.	0.28	2.26		0.28	0.68	n.s.	0.27	0.25	

[a] Group 1, Estraderm TTS 50; group 2, Estraderm TTS 50 + 0.4 mg progesterone vaginal suppository; group 3, 2 mg Progynova + 0.4 mg progesterone vaginal suppository; group 4, control group. $P \leq 0.05$, Statistically significant difference between before therapy and after 6 months (analyzed with a Wilcoxon test). n.s., not significant; S.D., standard deviation.

1, oestrogen (oestradiol) alone (Estraderm TTS® 50) ($n = 6$; age, 51–68 years; mean age, 58.7 years); group 2, oestrogen (oestradiol) transdermally and progesterone (Estraderm TTS® 50 and 0.4 mg progesterone vaginal suppository) ($n = 7$; age, 48–58 years; mean age, 54.4 years); group 3, oestrogen (oestradiol) orally and progesterone (2 mg Progynova® and 0.4 mg progesterone vaginal suppository) ($n = 8$, age, 45–59 years; mean age, 52.5 years); and group 4, without therapy, as a control group ($n = 3$; age, 50–65 years; mean age, 55 years). Due to the discontinuation of treatment by one patient in group 2 and by two patients in group 3, six patients remained in each group with HRT. Taking this fact into account, the mean time that had elapsed since the last menstrual period was 8.8 years (S.D., 10.9) in group 1, 8.2 years (S.D., 7.1) in group 2, 8.9 years (S.D., 6.8) in group 3 and 7.8 years (S.D., 4.6) in group 4. Two patients in group 1 had had previous HRT, while three women each in groups 2 and 3 and one patient in group 4 had a history of hormone replacement therapy. In all of these cases, however, HRT had been terminated at least half a year before.

The study was a longitudinal study with ad hoc data. Patients were randomly assigned to treatment groups based on the order of presentation. The female volunteers were rotationally assigned to groups 1, 2, 3 or 4. There were women who were not prepared to forego treatment and who were therefore assigned to the therapy group resulting from the randomization list. That is why the control group (group 4) consisted only of three test persons. To facilitate comparisons, all patients were treated between October and April; likewise, to exclude diurnal variations, measurements were performed at 14:00 h in all patients.

Dosing instructions were as follows: Estraderm TTS® 50 patches (Ciba-Geigy, Basel, Switzerland) given every 3–4 days; 2 mg Progynova® (Schering, Vienna, Austria), once daily; 0.4 mg progesterone vaginal suppositories, 10 days per month.

2.3. Measurement methods

Skin properties were measured by non-invasive methods to evaluate the effects during various systemic hormone replacement treatments. The following skin properties were measured monthly: skin surface lipids, epidermal skin moisture, skin elasticity and skin thickness (epidermis and dermis).

Measurements were performed under standardized conditions, i.e. a room temperature of 21°C and a humidity level of 40–45%. Prior to the measurements, patients were given 2 h to adapt to room conditions without covering the measurement sites by clothes. Patients were instructed not to use cosmetics on examination days. The measurement sites were selected to include both UV-exposed and non-exposed sites. The measurements were always performed by the same investigator. Mean values of multiple measurements were determined to avoid measuring inaccuracies. Women were advised not to change their cosmetic regimen (emollients) 3 months prior to and during HRT. Cosmetic compounds with anti-ageing effects (such as retinoic acid, glycolic acid, tretinoin, ascorbic acid or hormonal compounds) were not allowed.

The Sebumeter® SM 810 (Courage + Khazaka Electronic GMBH, Cologne, Germany) was used for quantitative measurements of skin surface lipids composed of sebum and corneal lipids. The device consists of a fat-stain photometer that measures the level of light transmission of a plastic sheet coated with sebum. The method is insensitive to humidity. A probe is pressed on to the skin region under investigation for 30 s at a constant pressure of 9.4 N/cm^2. The Sebumeter measures the variation of light transmission through the strip. The change in sheet transparency is computed and the result displayed in units that can then be converted into micrograms per square centimetre [12,13]. The variation of light transmission is proportional to the quantity of lipids absorbed. Three sites were studied: forehead, inner side of the right upper arm, and the suprasternal region — all of them were kept free from garments prior to the measurements.

The Corneometer® CM 820 (Courage + Khazaka Electronic GMBH, Cologne, Germany) determines the humidity level of the stratum corneum by measuring electrical capacitance. Alterations of epidermal skin hydration result in a change in

capacitance of the measuring condensator. The probe is applied to the skin for 1 s at a pressure of 7.1 N/cm^2. The degree of epidermal skin humidity is indicated in system-specific units [9,12–14]. One unit represents a water content of stratum corneum of 0.02 mg/cm^2, according to a measuring depth of 20 nm. Measurements were performed on three sites: left temporal bone, inner side of the left upper arm, and the suprasternal region.

Dermaflex A® (Cortex Technology ApS, Hadsund, Denmark) is a device for the rheologic measurement of skin elasticity based on a vacuum-induced elevation of skin, which is represented by the following factors: tensile distensibility (measured as the maximum skin elevation after one suction (mm)), elasticity (measured and calculated as the degree of total retraction after one suction (%)) and hysteresis (measured as the increase in maximum skin elevation in the last suction minus the distensibility (mm)). The measurement data depend on the composition of elastic and non-elastic structures of the skin. The main criterion is elasticity, which is defined as: (tensile distensibility-resilient distension) × 100%/tensile distensibility. Tensile distensibility reflects the collagen fibres, and resilient distension (the resultant elevation of the skin after release of the first suction) reflects the elastic fibres. The hysteresis parameter reflects the water content, the dominating viscous component of skin rheology, which determines the skin turgor. The measurements were performed as follows: magnitude of suction in the probe with a 150 mbar vacuum; number of suctions in the measurement preselected at 6 times, with each measuring cycle consisting of a suction and release phase lasting 6 s. Measurements were performed at the right mandibular region, the inner side of the right upper arm, and the suprasternal region.

Osteoson® D III (Minhorst GmbH & Co, Meudt, Germany) is a high-frequency ultrasound system for the measurement of skin thickness (epidermis and dermis) [12,15]. The probe has a frequency of 25 MHz and a bandwidth of 8 MHz, and measures the skin at a depth of up to 5 mm. The results are imaged both in an A-scan and a B-scan mode. The A-scan units represent the received ultrasound echo signals as a one-dimensional amplitude diagram displayed against time. All two-dimensional procedures are called B-scan procedures [16]. For the purpose of the present study, the distance between the first two tension amplitudes, i.e. between the interface of air-exposed epidermis and the dermal-subcutaneous fat tissue layer, was chosen [15]. An echogenic band caused by the entry echo (or epidermis/gel interface) is representing the epidermis. Due to limited axial resolution, normal epidermis cannot always be viewed separately. That is why the total thickness of the epidermis and dermis was measured [17]. Ultrasound was chosen to measure skin thickness, given that it is a highly precise method for this purpose [18]. Measurements were performed at the inner side of the left upper arm in order to exclude UV-induced influences on skin ageing.

In addition, clinical evaluation was performed at monthly intervals. This consisted of a questionnaire for climacteric complaints (Klimax Score) and for skin symptoms. The Klimax Score includes the following parameters: 1, psychovegetative symptoms: hot flushes, disturbed sleep, nervousness, fatigue, depression, libido reduction, palpitation, paresthesiae, dizziness, forgetfulness, headache; 2, atrophical symptoms: vaginal dryness, incontinence, arthralgia, myalgia, skin dryness, dry eyes; 3, progestagen-deficiency symptoms: breast tenderness, migraine, oedema. The intensity of symptoms was classified as follows: 0 = none, 1 = slight, 2 = moderate, 3 = distinct. Moreover, each patient's skin was monitored clinically according to a personal evaluation scheme at monthly intervals for the following criteria: skin dryness, skin tension, desquamation, slackness of skin, turgor, firmness, elasticity, moisture, and skin lipids. For this, the following evaluation scheme was used: (–) none, (+) minor, (+ +) moderate, (+ + +) marked and (+ + + +) very marked change.

Blood tests for oestradiol, follicle stimulating hormone and progesterone were performed by routine radioimmunoassay methods [19] before therapy and at 3-monthly intervals. A gynaecological examination and bone densitometry were performed before therapy.

Bone density screening was performed using a Hologic QDR-1000™ X-ray bone densitometer (Hologic, USA) based on the DXA measuring method. Sites of measurements were lumbar vertebrae [1–4]. Only patients with bone density values within the normal range were included in the study.

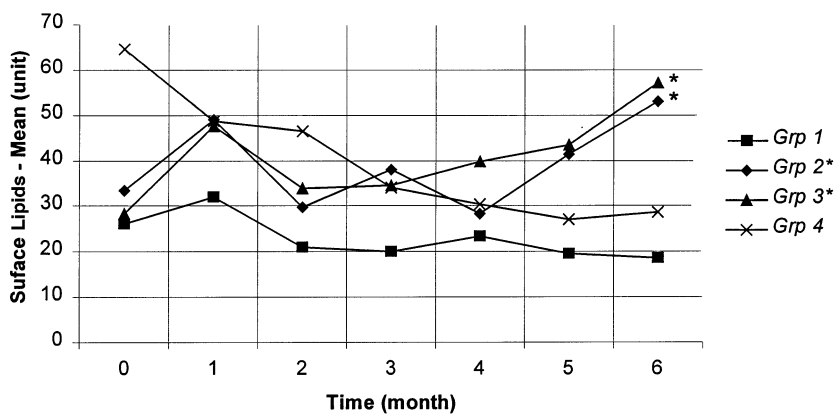
2.4. Statistical methods

The data were analysed using the SPSS (Superior Performing Software Systems) for Windows Release 8.00 (1997) program. Due to the small sample size, a normal distribution could not be assumed. A parametric test could not be applied either. Therefore, the Wilcoxon signed-rank test was used instead of a parametric test. For comparisons between groups, the H-test by Kruskal–Wallis was used, which compared the groups in a multivariant mode for significant differences. $P \leq 0.05$ was considered significant and $P \leq 0.01$ was considered highly significant.

3. Results

3.1. Skin surface lipids

When only oestrogen was given (i.e. without progesterone), we observed a significant decrease in skin lipids (except in the suprasternal region where the decrease was not significant). The largest decrease was found at the inner side of the upper arm. In contrast, the combination of oestrogen plus progesterone resulted in a significant increase in skin surface lipids. The largest increase was detected in the group receiving 2 mg Progynova plus 0.4 mg progesterone vaginal suppository. The control group showed no significant changes (Table 2 and Fig. 1). The difference in



Grp1: *Estraderm TTS 50 R*

Grp2: *Estraderm TTS 50R & progesterone vaginal supp. 0,4 mg*

Grp3: *Progynova R 2 mg & progesterone vaginal supp. 0,4 mg*

Grp4: *Control group*

** p ≤ 0.05: statistically significant difference between before therapy and after 6 months.*

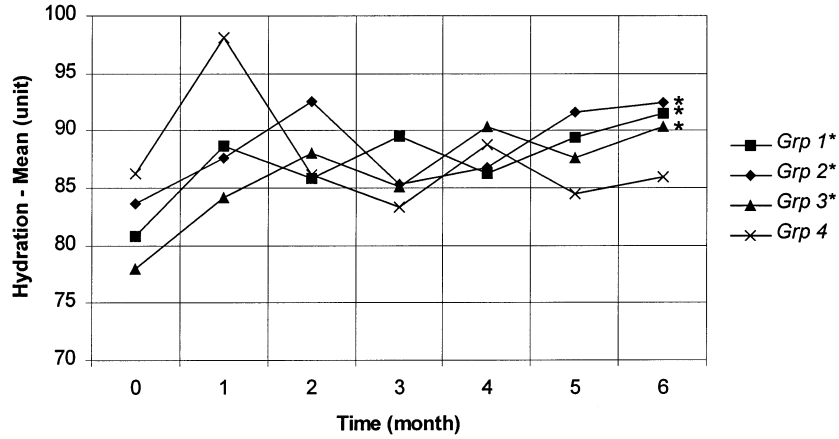
(analyzed with a Wilcoxon test)

Fig. 1. Skin surface lipids: suprasternal.

Table 2
Measurements[a]

		Group 1			Group 2			Group 3			Group 4		
		Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*
Sebumetry (units)													
Frontal	Mean	154.33	133.83	0.03	129.83	163.00	0.03	152.67	210.67	0.03	111.33	119.67	n.s.
	S.D.	44.27	43.68		42.46	42.31		57.44	46.39		37.17	13.43	
Inner side of the	Mean	43.83	5.33	0.04	1.83	5.00	0.04	2.00	10.33	0.04	11.00	5.00	n.s.
Right upper arm	S.D.	76.09	7.28		0.75	2.10		1.79	8.73		7.21	5.20	
Suprasternal	Mean	26.17	18.67	0.22	33.33	53.00	0.05	28.33	57.17	0.03	64.67	28.67	n.s.
Region	S.D.	22.13	13.62	n.s.	23.00	23.10		12.34	17.08		39.80	19.40	
Corneometry (units)													
Frontal	Mean	72.22	86.90	0.03	69.22	84.28	0.03	71.35	82.45	0.03	63.00	73.00	n.s.
	S.D.	7.98	3.60		12.36	6.49		9.23	9.85		7.05	3.38	
Inner side of the	Mean	61.67	85.06	0.03	63.55	78.48	0.03	64.05	75.38	0.03	73.37	70.53	n.s.
Left upper arm	S.D.	2.35	6.32		9.31	6.13		10.18	8.46		4.73	5.69	
Suprasternal	Mean	80.85	91.55	0.03	83.68	92.48	0.05	77.95	90.40	0.03	86.23	85.90	n.s.
Region	S.D.	9.25	5.70		5.84	2.39		11.97	5.09		5.28	4.16	
Elasticity (%)													
Mandibular region	Mean	51.67	66.67	0.03	42.17	61.17	0.03	50.67	67.50	0.03	54.67	48.33	n.s.
Right side	S.D.	11.41	8.94		11.87	12.53		7.00	9.87		10.97	6.81	
Inner side of the	Mean	34.33	41.50	0.03	36.50	45.50	0.03	39.67	55.83	0.03	56.00	39.00	n.s.
Right upper arm	S.D.	5.24	5.54		3.27	5.17		8.16	5.71		18.25	10.15	
Suprasternal	Mean	41.17	62.33	0.03	49.67	59.50	0.03	45.33	65.50	0.03	64.33	57.67	n.s.
Region	S.D.	9.41	13.71		9.03	5.17		9.73	11.38		22.14	15.28	
Skin thickness (mm)													
Inner side of the	Mean	0.906	1.118	0.03	0.900	1.030	0.03	0.891	1.046	0.03	0.848	0.881	n.s.
Left upper arm	S.D.	0.154	0.124		0.099	0.097		0.094	0.065		0.106		

[a] Group 1, Estraderm TTS 50; group 2, Estraderm TTS 50+0.4 mg progestrone vaginal suppository;group 3, 2 mg Progynova+0.4 mg progesterone vaginal suppository; group 4, control group. $P \leq 0.05$, Statistically significant difference between before therapy and after 6 months (analyzed with a Wilcoxon test). n.s., Not significant; S.D., standard deviation.



Grp 1: *Estraderm TTS 50 R*

Grp2: *Estraderm TTS 50R & progesterone vaginal supp. 0,4 mg*

Grp3: *Progynova R 2 mg & progesterone vaginal supp. 0,4 mg*

Grp4: *Control group*

** p ≤ 0.05: statistically significant difference between before therapy and after 6 months.*

(analyzed with a Wilcoxon test)

Fig. 2. Epidermal hydration: suprasternal.

baseline values between group 4 and the other groups can be seen from the descriptive data. It is not measurable significantly by statistical methods, however, on account of the small sample size in group 4.

3.2. Epidermal hydration

Epidermal moisture showed comparable significant increases in all HRT groups after 6 months of treatment at all measuring sites. The increase was comparable in all HRT groups for all measuring sites (Fig. 2). The largest increase in hydration was observed at the inner side of the upper arm in patients taking only oestrogen. There were no significant changes in the control group.

3.3. Skin elasticity

Skin elasticity was significantly improved after 6 months of HRT. The increases were smaller at the inner side of the upper arm than at the mandibular region and the suprasternal region (Fig. 3). The changes in the control group were not significant.

A comparison of the measurements of epidermal hydration and skin elasticity revealed no significant differences between UV-exposed and non-exposed measurement sites.

3.4. Skin thickness

Skin thickness increased significantly in all hormonally treated groups as compared with the control group. The mean value of skin thickness in all HRT groups increased at a highly significant level of 0.17 mm after 6 months of HRT ($P = 0.0002$) (Fig. 4).

3.5. Clinical efficacy

The Klimax Score demonstrated significant improvements of clinical symptoms in all hormonally treated groups.

Clinically, improvements of skin dryness, skin tension, desquamation, slackness of the skin, turgor, firmness, elasticity and moisture were observed with HRT. In patients receiving HRT, a reduction of skin lipids was observed after 6 months, except for the Estraderm TTS® 50 and 0.4 mg progesterone vaginal suppository group (Table 3).

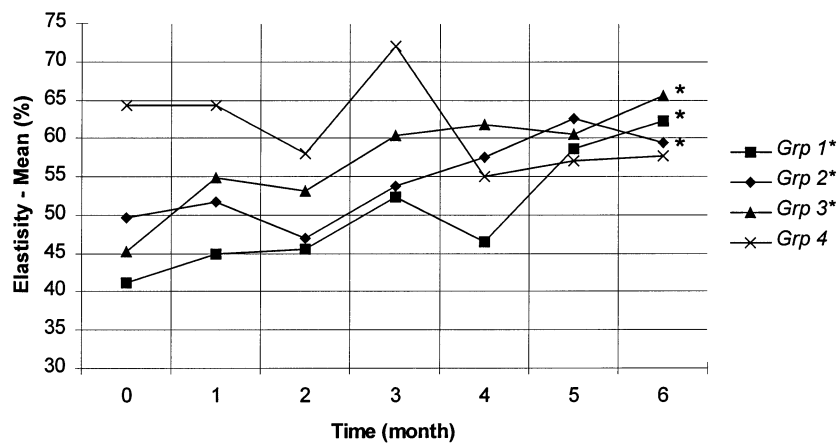
In most cases, the largest increases in subjective and clinical improvements were observed between the third and fourth months of treatment. A clinically significant improvement of skin dryness and elasticity became evident at the end of treatment.

3.6. Hormone levels

The evaluation of hormone serum levels (serum oestradiol, follicle stimulating hormone and progesterone) showed increases in oestradiol and progesterone and decreases in FSH in all treatment groups during HRT (Table 1). The results were not significant with the exception of the rise in progesterone in group 2.

3.7. Adverse effects

In group 1, three patients complained of temporary breast tenderness, and one of local reddening at the application site of the hormone patches. One patient each complained about transient breast tenderness in groups 2 and 3. Local reddening caused by the patch was observed in group 2.



Grp1: *Estraderm TTS 50 R*

Grp2: *Estraderm TTS 50R & progesterone vaginal supp. 0,4 mg*

Grp3: *Progynova R 2 mg & progesterone vaginal supp. 0,4 mg*

Grp4: *Control group*

** p ≤ 0.05: statistically significant difference between before therapy and after 6 months.*

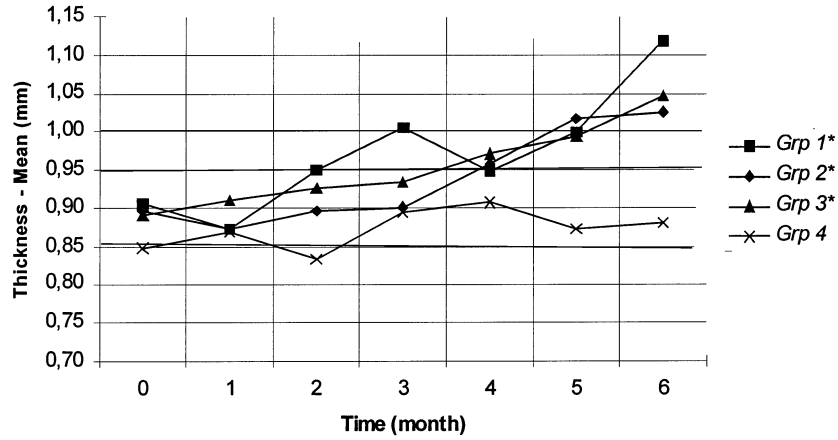
(analyzed with a Wilcoxon test)

Fig. 3. Skin elasticity: suprasternal.

Table 3
Clinical changes[a]

		Group 1			Group 2			Group 3			Group 4		
		Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*	Before therapy	After 6 months	*P*
1 Skin dryness	Mean	3.67	1.25	0.03	2.75	1.17	0.03	2.67	0.58	0.03	3.67	3.00	n.s.
	S.D.	0.82	1.29		1.33	1.13		0.52	0.49		0.58	1.73	
2 Skin tension	Mean	3.00	0.75	0.03	2.17	0.75	0.04	2.33	0.50	0.03	3.00	2.67	n.s.
	S.D.	1.10	1.17		1.60	1.17		1.21	0.55		0.00	1.53	
3 Desquamation	Mean	1.67	0.17	0.04	0.83	0.33	0.08	1.33	0.00	0.04	1.67	1.33	n.s.
	S.D.	1.86	0.41		1.17	0.82	n.s.	0.82	0.00		1.53	1.53	
4 Slackness of the skin	Mean	2.50	1.25	0.07	2.42	1.50	0.07	2.50	0.92	0.04	2.00	2.17	n.s.
	S.D.	1.38	1.08	n.s.	1.36	1.00	n.s.	0.55	0.66		0.00	1.76	
5 Turgor	Mean	1.50	2.75	0.02	1.67	2.67	0.07	2.00	3.17	0.06	2.33	2.00	n.s.
	S.D.	0.55	0.88		0.82	0.52	n.s.	0.63	0.98	n.s.	0.58	1.00	
6 Firmness	Mean	1.50	2.75	0.02	1.67	2.83	0.07	2.00	3.33	0.07	1.67	1.67	n.s.
	S.D.	0.55	0.88		0.82	0.75	n.s.	0.63	1.03	n.s.	0.58	0.58	
7 Elasticity	Mean	2.00	3.67	0.41	1.83	3.00	0.04	2.17	3.33	0.07	1.67	1.67	n.s.
	S.D.	0.89	0.52	n.s.	0.75	0.89		0.75	0.82	n.s.	0.58	1.15	
8 Moisture	Mean	0.17	1.00	0.10	1.00	2.17	0.04	1.17	2.33	0.10	0.67	2.00	n.s.
	S.D.	0.41	0.89	n.s.	1.10	1.17		0.75	1.63	n.s.	0.58	1.00	
9 Skin lipids	Mean	1.50	0.83	0.10	1.00	1.00	1.00	2.17	1.67	0.41	1.00	11.00	n.s.
	S.D.	1.22	0.75	n.s.	1.26	1.10	n.s.	1.33	1.21		1.73	19.05	

[a] Group 1, *Estraderm TTS 50;* group 2, Estraderm TTS 50+0.4 mg progesterone vaginal suppository; group 3, *2 mg Progynova+0.4 mg progesterone vaginal suppository; group* 4, *control group*. 0, No change; 1, minor change; 2, moderate change; 3, marked change; 4, very marked change. $P \leq 0.05$, Statistically significant difference between before therapy and after 6 months (analyzed with a Wilcoxon test). n.s., Not significant; SD, standard deviation.



Grp1: *Estraderm TTS 50®*

Grp2: *Estraderm TTS 50® & progesterone vaginal supp. 0,4 mg*

Grp3: *Progynova® 2 mg & progesterone vaginal supp. 0,4 mg*

Grp4: *Control group*

** $p \leq 0.05$: statistically significant difference between before therapy and after 6 months.*

(analyzed with a Wilcoxon test)

Fig. 4. Skin thickness: upper arm inside.

4. Discussion

In the present study, the clinical finding of beneficial HRT effects on skin ageing symptoms were substantiated by objective measurements, whereas we did not detect any significant changes in the control group.

The finding of increased skin surface lipids during combined HRT may reflect stimulatory effects of the progestagen component on sebaceous gland activity, in contrast to oestrogen alone having a sebum-suppressive activity [20–22].

The amount of sebum on the skin surface reflects the size and density of the sebaceous glands. It is largest on the forehead and decreases in the following order: chest, back, abdomen, upper and lower limbs [23]. An increase in sebum secretion with HRT (oestradiol plus various progestagens) has been previously reported by Callens et al. [12] and Pierard [22]. However, a distinct influence of progestagens on skin ageing has not been described [8]. Our findings suggest that a decrease in sebum production during menopause may be alleviated by a course of combined HRT (oestrogen plus progesterone) [24]. This might be relevant to atopic patients, who might benefit from increased epidermal lipids. It is also important to note that hormonally increased sebum production may be of relevance in acne-prone skin, and that this may be the reason for acne flares in predisposed females undergoing HRT. Although acne flares were not observed in our study, skin lipid increases indicate such a possibility and it is imperative for hormone substitution to be carefully adapted to each patient's specific situation.

An additional aspect of hormonal effects on connective tissue was obtained by measuring skin elasticity. Comparable significant increases in

elasticity were observed in all treatment groups by month 6 of HRT, which is indicative of the positive effects of various kinds of HRT on tensile skin functions.

In addition, our study demonstrates significant improvements after 6 months of treatment not only of elasticity, but also of epidermal hydration in all patients receiving HRT, a finding which is in agreement with clinical observations by Dunn et al. [25]. In contrast, Callens et al [12] and Jemec and Serup [26] found no modification of superficial skin hydration in patients receiving HRT.

The lack of significant differences between the increase in epidermal hydration and elasticity during HRT in UV-exposed and non-exposed sites of the skin is of great importance because it suggests that, in certain aspects, photoaged skin may benefit from hormonal treatment in a similar way as UV-protected aged skin. Our results are in accordance with the study of Henry et al. [27].

Epidermal and dermal thickness, the structure of elastic fibres, the skin vessels and the dermal mucopolysaccharide acid content are important markers for the progression of skin ageing. All these parameters are under oestrogen influence [9].

Our results show a highly significant increase in skin thickness (epidermal and dermal thickness) in hormonally treated postmenopausal women after a 6-month treatment period. In addition, our clinical observations suggest that HRT (particularly in patients with 2 mg Progynova® with 0.4 mg progesterone vaginal suppository) may diminish teleangiectasia, which were less visible, possibly due to increased epidermal thickness.

A leading parameter of skin ageing is skin thickness, which reflects the status of the collagen and elastic tissue. Punnonen and Rauramo [28] showed significant increases in both skin collagen and thickness during HRT in a histological investigation. Their findings were supported by Brincat et al. [29], who showed a positive correlation between bone collagen and skin collagen, both decreasing after menopause and increasing during HRT. Using ultrasound, the authors demonstrated an increase in epidermal and dermal thickness a few weeks after the start of systemic oestrogen substitution therapy. Moreover, a recent study demonstrated the action of oestrogen on dermal components such as collagen, elastic fibres, water content, hyaluronic acid and fibroblasts [12].

Increases in skin thickness were also reported by other authors who used ultrasound [7,12,15] and other measuring techniques [29,30]. Our study, in which ultrasound, a sensitive, reproducible and valid technique for measurements of skin thickness [15,18], was used, supports these results. In contrast, one large cross-sectional study reported that oestrogen use was associated with thinner skin (by calibre assessment) [31].

The clinical improvements of the various skin properties during HRT were in accordance with the measurement data. Especially with oestrogen alone, the use of cosmetic creams was reduced, although the skin surface lipids decreased. This finding confirms the increase in epidermal hydration during oestrogen treatment. Other positive findings noted by the patients were a moderate reduction of wrinkles and an increased firmness of the skin at the end of the treatment period.

There were only minor side effects of HRT and they consisted of breast tenderness, and of local reddening at the application site of the hormone patches in some of the patients.

HRT was shown to significantly improve parameters involved in skin ageing in this preliminary study. In most cases, hydration, elasticity and skin thickness were significantly increased. These findings support the clinical impression that an early hormone substitution therapy may restore the initial features of hormonally induced skin ageing. Whether HRT will be able to delay the onset of skin ageing in general or to alleviate the severity of its symptoms will be subject to further research.

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