A Study of the Decrease of Skin Collagen Content, Skin Thickness, and Bone Mass in the Postmenopausal Woman

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The skin collagen content, skin thickness, metacarpal index, and forearm bone mineral content in postmenopausal women showed a similar decline of between 1–2% per year after the menopause. All four parameters showed a decline that was significant when compared with the years from the menopause. Significant correlations between all four parameters suggest that a similar pathology causes the decrease in bone mass and skin thickness—a decline in the connective tissue element that is common to both bone and skin. ***(Obstet Gynecol 70:840, 1987)***

Collagen constitutes approximately one-third of the total mass of the body.[1] Skin and bone share a similar loose connective tissue in the dermis and the organic matrix. The predominant collagen of bone is type I.[2] The dermis is also largely composed of type I collagen, although type III is also present.

There is evidence that skin collagen is affected by hypoestrogenism[3] and decreases in the years after the menopause.[4] Skin thickness measurements largely indicate dermal thickness, with the epidermis accounting for only 7% of the total thickness.[5] These measurements can be used as a model for studying the effects of sex steroids on connective tissue. Skin thickness has also been shown to be affected with estrogen therapy and with postmenopausal hypoestrogenism.[4] Skin thickness measurements represent changes in dermal connective tissue and amorphous ground substance.

Connective tissue greatly contributes to the strength of the bone; bone without collagen is brittle, like chalk.[6] The possibility must be considered that the decline that hypoestrogenism causes in bone mass, skin collagen, and skin thickness may have a common etiology. This would lead to thin skin, with a low collagen content, and to osteoporotic bones. This study was carried out to study the relationship and rate of decline of four parameters, skin collagen content, skin thickness, metacarpal index, and bone mineral content in the years after the menopause.

Materials and Methods

One hundred forty-eight women attending the Dulwich Hospital Menopause clinic for the first time, and who had not been on any form of sex hormone treatment since their menopause, were asked to participate in the study. Skin biopsy specimens were taken from all patients.

One hundred thirty-three of these patients were recruited for the skin thickness studies, and had a skin x-ray and a hand x-ray taken. The 68 patients recruited for the bone mineral content studies also had a bone mineral content assessment. Women were entered into these studies sequentially; thus the earliest and first group of women to be recruited had skin biopsies only. Those recruited later also had skin thickness and hand x-rays, and the latest group recruited had a bone mineral content assessment in addition to the x-rays and the skin biopsy.

Table 1 presents the patient data for all these groups of women.

Duration of the menopause was determined from the last menstrual period, or in cases of women who had undergone a hysterectomy only, from the date of onset of characteristic symptoms of the climacteric.[7] Women who had a bilateral oophorectomy in addition to their hysterectomy were considered menopausal from the date of operation. Serum follicle-stimulating hormone (FSH) was also measured; a patient was deemed postmenopausal if her FSH was high.[8]

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Supported by a Birthright Grant from the Royal College of Obstetricians and Gynaecologists.

Table 1. Patient Data of the Women Used in Studies of Thigh Skin Collagen, Forearm Skin Thickness, Metacarpal Index, and Bone Mineral Content

Data	SC	ST	MI	BMC
N	148	133	128	68
Age (yr)	51 ± 7.9	51.1 ± 8.7	51.1 ± 8.8	53 ± 7.7
Menopausal age (yr)	5.4 ± 6.5	5.9 ± 7.0	5.9 ± 6.3	6.4 ± 7.3
Weight (kg)	65 ± 11	64.6 ± 12	65.2 ± 11.3	68.4 ± 11.1
Height (m)	1.61 ± 0.07	1.61 ± 0.07	1.61 ± 0.08	1.61 ± 0.06

SC = skin collagen; ST = skin thickness; MI = metacarpal index; BMC = bone mineral content.
Data are expressed as mean ± SD.

For the measurement of skin collagen content, punch biopsy specimens of skin 3 mm in diameter were taken from the right thigh, 5 cm below the greater trochanter. Hydroxyproline was extracted from these specimens and measured using Woessner's method.[9] The collagen content was then calculated as described by Neuman and Logan.[10] These authors showed that most of the hydroxyproline in the skin was of collagenous origin, with a minimal amount derived from elastin. The total error of the method was 11.4% in our laboratory; this figure includes the interassay and intra-assay variations. The sensitivity of the assay, especially at the range used, was high. The results were expressed in relation to the surface area of the skin biopsy ($\mu g/mm^2$).[11]

No attempts were made in this study to investigate the individual types of collagen. At this stage, we looked only at the total skin collagen.

The skin thickness on the ulnar aspect of the forearm at a constant distance from the elbow was measured using the x-ray frame shown in Figure 1. The frame consisted of a mahogany rectangular block, 4 cm long and 15 mm wide, pressing against the ulnar aspect of the forearm with the central beam directed at 90° horizontally against the cross-section of the skin. This design eliminated the blurring of the edges at the top of the epidermis and the bottom of the dermis caused by the convexity of the arm. Mahogany is radiotranslucent, like subcutaneous tissue. Because skin is not radiotranslucent, compression of the skin with a wooden block makes the top of the epidermis and the

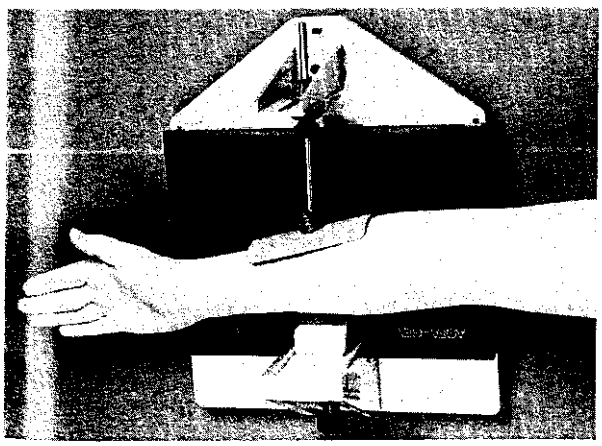

Figure 1. Apparatus for measuring skin thickness radiographically. The x-ray plate is placed behind the forearm and the x-ray is taken from above.

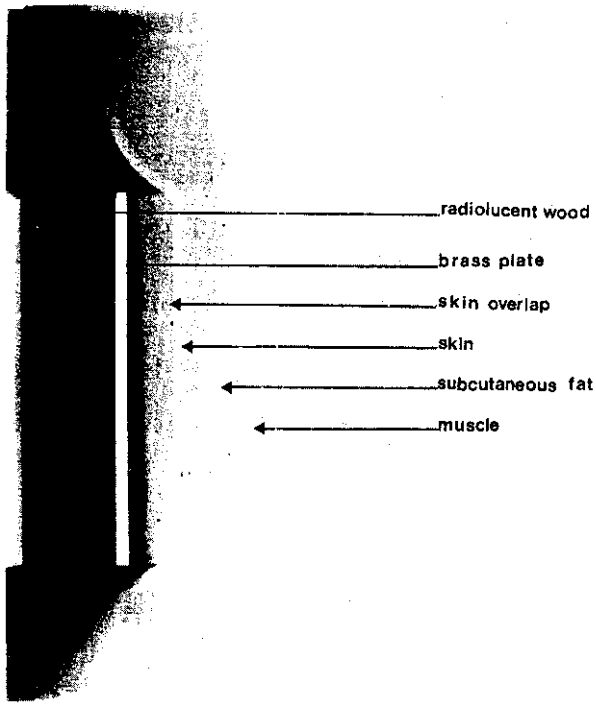

Figure 2. X-ray of the skin thickness.

Table 2. Thigh Skin Collagen Content ($\mu g/mm^2$) in 148 Untreated Postmenopausal Women With Years Since Menopause

MA	*N*	Mean ± SD
0–0.25	19	189.6 ± 55.2
0.5–0.6	14	193.4 ± 55.6
1	19	161.1 ± 44.8
1.5–2	16	191.7 ± 73.5
3	14	192.4 ± 66.9
4–5	15	146.6 ± 54.0
6–7	13	147.1 ± 57.7
8–9	15	154.5 ± 81.3
10–15	12	130.6 ± 30.6
16+	11	108.6 ± 23.3

MA = years since menopause.

Figure 3. X-ray of the second metacarpal. The same x-ray plate is used to assess skin thickness and the second metacarpal, thus enabling the metacarpal index to be assessed.

bottom of the dermis clearly visible. The skin appears as a thick radio-opaque band between two parallel lines, gently compressed between the radiotranslucent wooden block and the subcutaneous tissue (Figure 2). A 1-mm brass plate was placed in the wooden block 0.5 mm from the functional edge. This showed sharply on the x-ray films and was used as a control to correct the skin thickness measurements.

After the exposure of the forearm skin, we made a second x-ray exposure of the second metacarpal of the right hand by positioning the hand in the posteroanterior position on another part of the same film and coning the x-ray beam to cover the second metacarpal only. This technique gave a higher resolution and made the edges more easily identifiable. Both skin thickness and metacarpal index were therefore assessed on the same film (Figures 2 and 3). The same observer measured all the skin thickness and metacarpal x-rays.

Measurements were carried out using Vernier calipers. The metacarpal index was the cortical area (total area ratio CA/TA) calculated using the formula $CA/TA = 1 - (1-C/T)^2$, where C = cortical thickness and T = total diameter.[12]

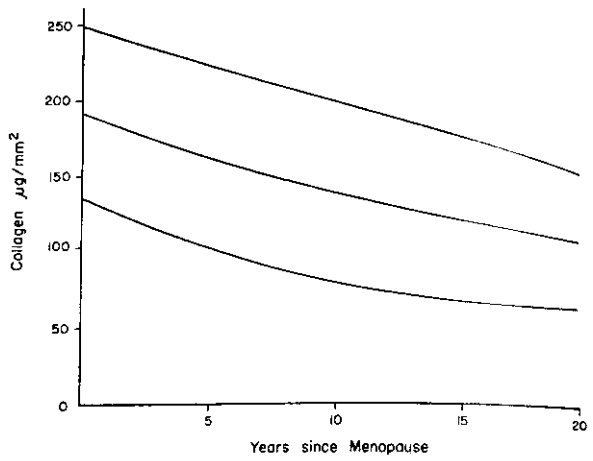

Figure 4. The mean ± SD thigh skin collagen content of 148 postmenopausal women plotted against years since the menopause. There is a highly significant negative correlation.

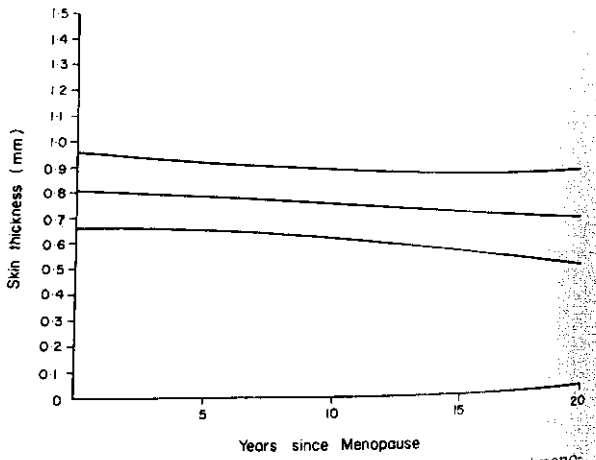

Figure 5. The mean ± SD forearm skin thickness in 133 postmenopausal women plotted against years since the menopause. There is a highly significant negative correlation.

Table 3.

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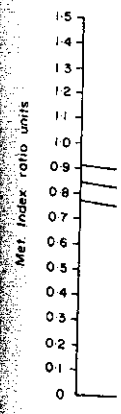

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Table 3. Skin Thickness (mm) in 133 Untreated Postmenopausal Women at Various Menopausal Age Ranges

MA	N	Mean ± SD
0–0.25	12	0.88 ± 0.14
0.5–0.6	10	0.77 ± 0.15
1	20	0.75 ± 0.16
1.5–2	17	0.81 ± 0.13
3	9	0.80 ± 0.14
4–5	15	0.77 ± 0.14
6	14	0.73 ± 0.12
7–10	12	0.81 ± 0.12
11–19	12	0.69 ± 0.14
20+	12	0.64 ± 0.12

MA = years since menopause.

Table 4. Metacarpal Index With Menopausal Age Ranges in 128 Untreated Postmenopausal Women

MA	N	Mean ± SD
0–0.25	12	0.85 ± 0.05
0.5–0.6	8	0.81 ± 0.10
1	19	0.82 ± 0.08
1.5–2	17	0.81 ± 0.06
3	8	0.82 ± 0.08
4–5	14	0.84 ± 0.06
6	14	0.79 ± 0.08
7–10	12	0.79 ± 0.08
11–19	12	0.67 ± 0.10
20+	12	0.69 ± 0.12

MA = years since menopause.

To measure bone mineral content, we used peripheral single (beam) photon absorptiometry using one isotope. A Gambro bone mineral detector was used, and readings were taken from the left forearm at 3 and 8 cm from the epicondyles. Two readings were taken from the radius and two from the ulna, and the mean bone mineral content of the two bones was calculated.

Correlations were calculated using the Pearson's correlation test (r). Curves were fitted when necessary, using the least mean squares difference[13] on a Hewlett Packard Program. The distribution of the data was shown to be normal using the Kolmogorov-Smirnov one-sample test[14] available on the SPSS-X program.

Results

The thigh skin collagen decreased with the years since the menopause from a mean of 190 $\mu g/mm^2$ at the start of the menopause to a mean of 130 $\mu g/mm^2$ ten to 15 years after the menopause. There was no observed relationship between thigh skin collagen content and actual chronological age (Figure 4, Tables 2 and 6).

There was also a relationship between skin thickness and years since menopause. The skin became thinner with increasing menopausal age, from a mean of 0.88 mm at the start of the menopause to a mean of 0.69 mm at ten years after the menopause. There was no relationship found between skin thickness and chronological age (Figure 5, Tables 3 and 6).

We found a relationship between metacarpal index and both the years since the menopause and the chronological age of the patient. The metacarpal index declined with increasing age and increasing menopausal age (Figure 6, Tables 4 and 6) from a mean of 0.85 at the start of the menopause to 0.67 at 11–19 years after the menopause.

Five observations at 8 cm and nine observations at 3 cm were invalid because of technical errors, such as

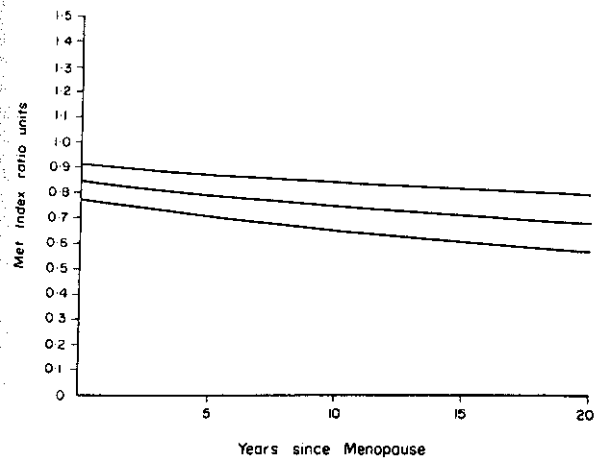

Figure 6. The mean ± SD metacarpal index (CA/TA) in 128 postmenopausal women plotted against years since the menopause. There is a highly significant negative correlation.

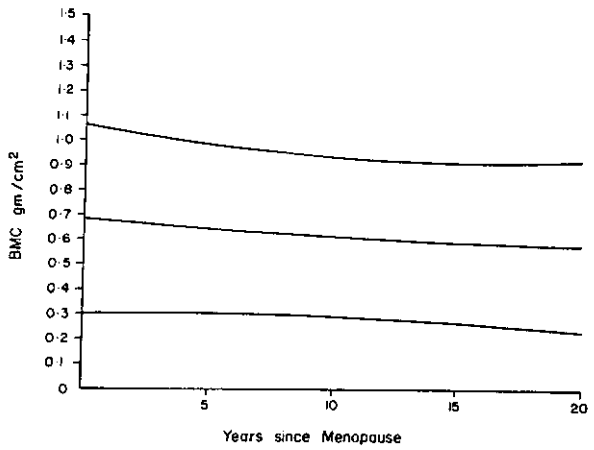

Figure 7. The mean ± SD bone mineral content at 8 cm up the forearm in 63 postmenopausal women plotted against years since the menopause. There is a significant negative correlation.

Table 5. Bone Mineral Content (g/cm^2) at 3 and 8 cm

	3 cm		8 cm	
MA	*N*	Mean ± SD	*N*	Mean ± SD
0.06	10	0.47 ± 0.11	7	0.71 ± 0.13
1–1.5	0	0.49 ± 0.10	10	0.67 ± 0.12
2–3	8	0.45 ± 0.11	12	0.60 ± 0.06
4–5	9	0.46 ± 0.07	10	0.66 ± 0.09
6–8	11	0.45 ± 0.06	12	0.64 ± 0.04
10+	11	0.45 ± 0.08	12	0.56 ± 0.11

MA = years since menopause.

Table 7. Correlations Between Various Parameters in Postmenopausal Women

	MI			SC			ST		
Parameter	*N*	*r*	*P*	*N*	*r*	*P*	*N*	*r*	*P*
MI				128	0.37	<.0001	124	0.41	<.0001
BMC 3 cm	59	0.38	<.001	59		NS	59		NS
BMC 8 cm	63	0.41	<.0001	63	0.24	<.05	63	0.24	<.05
ST	124	0.41	<.0001	109	0.45	<.0001			

MI = metacarpal index; SC = thigh skin collagen content; ST = forearm skin thickness; BMC 3 cm = bone mineral content 3 cm up the forearm from the styloid process; NS = not significant; BMC 8 cm = bone mineral content 8 cm up the forearm from the styloid process.

involuntary arm movements, when the measurements were taken.

The bone mineral content at 3 cm up the forearm (primarily cortical bone, but with some trabecular bone present[16]) showed no relationship with either chronological age or menopausal age. The bone mineral content at 8 cm (predominantly cortical bone[16]) had a highly significant correlation with both chronological age and menopausal age ($P < .0001$ in both cases), with the bone mineral content decreasing with increasing menopausal age and increasing chronological age (Figure 7, Tables 5 and 6). The mean bone mineral content at 8 cm was 0.71 g/cm^2 at the start of the menopause, and dropped to 0.56 g/cm^2 at ten years after the menopause.

Table 6 presents the correlations obtained between skin collagen, skin thickness, metacarpal index, and bone mineral content at 3 cm and at 8 cm with years after the menopause and with chronological age. Comparisons among the five skin and bone density parameters (Table 7) indicate a relationship between them.

Discussion

Our cross-sectional data show a highly significant correlation between changes in thigh skin collagen, forearm skin thickness, metacarpal index, and the mean bone mineral content at 8 cm and the years since the menopause (Table 6). No correlation was found between bone mineral content at 3 cm and years after the menopause. At this 3-cm level, measurements using the photonabsorptiometer are less accurate, and the bone, although primarily cortical, has some trabecular bone present.[15] At 8 cm, there is predominantly cortical bone, which is similar to that present in the second metacarpal.[15]

Although the decrease in skin collagen, skin thickness, metacarpal index, and bone mineral content at 8 cm is not linear, an annual linear decline with years since the menopause can be calculated. Thus, the average linear decline of skin collagen is 2.1% per postmenopausal year up to 15 years after the menopause. The average annual linear decline in skin thickness in the first 19 years after the menopause is 1.13% per postmenopausal year, virtually identical to the annual linear decline of 1.22% in metacarpal index in the first 19 years after the menopause. The average linear decline of bone mineral content at 8 cm in the first 11 postmenopausal years is 0.94% per postmenopausal year.

The average decline in metacarpal index that we obtained in this study is similar to the 10% per decade reported by Newton-John and Morgan in 1968.[16] Unlike skin collagen and skin thickness, both bone indices, metacarpal index and bone mineral content at 8 cm, had highly significant correlations with the chronological age of the woman. This implies that these

Table 6. Thigh Skin Collagen, Skin Thickness, Metacarpal Index, and Bone Mineral Content in Postmenopausal Women, and Correlations With Years Since Menopause and Chronological Age

			Correlation with menopausal age		Correlation with chronological age	
Parameter	*N*	Mean ± SD	*r*	*P*	*r*	*P*
Skin collagen ($\mu g/mm^2$)	148	165.4 ± 62	−0.30	<.0001	No correlation	
Skin thickness (mm)	133	0.76 ± 0.15	−0.33	<.0001	No correlation	
Metacarpal index	128	0.79 ± 0.1	−0.49	<.0001	−0.31	<.0001
BMC 3 cm (g/cm^2)	59	0.4625 ± 0.09	No correlation		No correlation	
BMC 8 cm (g/cm^2)	63	0.6314 ± 0.1	−0.4143	<.001	−0.4333	<.0001

BMC = bone mineral content.

parameters decline with increasing age, but also that the estrogen deficiency of the menopause adds to that decline.

The similar decline in all the parameters with years after the menopause suggests that there is an interrelationship between these parameters, with some common factor governing their similar response to hypoestrogenism. This common factor in skin collagen, skin thickness, and metacarpal index could be the connective tissue element present in all three. The decline in the bone mineral content at 8 cm is also related to the connective tissue element that supports it.

There are other conditions involving a connective tissue disorder of the skin and bones, notably osteogenesis imperfecta (brittle bone syndrome). In this genetic disorder, the abnormal collagen in both the skin and the bone is thought to be responsible for the worst manifestations of the disease.[17]

Our results indicate that dermal skin collagen and skin thickness decline after the menopause. Bone mass as shown in this cross-sectional data also declines after the menopause. Similar results have been shown in other prospective studies.[18–20] These findings alone would not necessarily mean that the two skin measurements share more than a similar susceptibility to hypoestrogenism. However, the data have also shown significant correlations between skin collagen and metacarpal index, and metacarpal index and bone mineral content at both 3 and at 8 cm.

Horsman and Kirby[20] have shown that metacarpal index is strongly representative of bone mass. Bone mass is composed of connective tissue (organic matrix, bone collagen) and mineral; the connective tissue element is also common to skin collagen, skin thickness, and metacarpal index. This connective tissue element varies from the skin to the bone. To support our hypothesis, definitive evidence is needed regarding the mechanism whereby estrogen deficiency affects the connective tissue in skin and bone. This study indicates that the hypoestrogenism occurring after the menopause has a profound effect on skin connective tissue and skin thickness, and contributes to their decline. Whether such a hypoestrogenism-mediated connective tissue defect also results in postmenopausal bone loss remains to be proved.

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Received December 8, 1986.
Received in revised form June 10, 1987.
Accepted June 10, 1987.