

Accuracy and reliability of a hand-held *in vivo* skin indentation device to assess skin elasticity

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Abstract

OBJECTIVE: The aim of this study was to evaluate the performance of a hand-held indentation device for fast and reliable determination of skin stiffness.

METHODS: Device accuracy to indentation depths of 0.6 and 1.3 mm was first evaluated on plastic foam materials with mechanical properties verified by a laboratory material testing device. Subsequently, the device's sensitivity to detect age-related changes in skin stiffness was evaluated among 46 healthy women (18–79 years). Finally, the reproducibility of the method was tested with six healthy subjects.

RESULTS: High correlation was detected between indentation stiffness of reference material and Young's modulus determined with mechanical testing device (0.6 mm indenter: $r = 0.97$, $P = 0.05$; 1.3 mm indenter: $r = 0.98$, $P = 0.04$). Age-related decrease of 38% in skin stiffness was observed in healthy volunteers ($P < 0.05$). The coefficient of variation for 0.6 and 1.3 mm indenters was 7.4% and 8.5%, respectively. No trend related to hysteresis effect was observed from repeated measurements.

CONCLUSIONS: The presented indentation technique was accurate against the laboratory material testing device. Furthermore, skin changes related to ageing could be detected with the indentation technique. The new device was found to be feasible for monitoring skin stiffness in cosmetics and clinical conditions.

Résumé

OBJECTIF: Cette étude avait pour objectif d'évaluer les performances d'un dispositif d'indentation portatif dans la détermination rapide et fiable de la rigidité de la peau.

MÉTHODES: La précision du dispositif à des profondeurs d'indentation de 0,6 et de 1,3 mm a d'abord été évaluée sur des matériaux de mousse plastique dont les propriétés mécaniques ont été vérifiées au moyen d'un dispositif d'essai des matériaux de laboratoire. La sensibilité du dispositif dans la détection des variations de la rigidité de la peau liées à l'âge a ensuite été évaluée chez 46 femmes en bonne santé (âgées de 18 à 79 ans). Enfin, la reproductibilité de la méthode a été testée sur six sujets en bonne santé.

RÉSULTATS: Une corrélation élevée a été détectée entre la rigidité d'indentation du matériau de référence et le module de Young

déterminé au moyen d'un dispositif d'essai mécanique (pénétrateur de 0,6 mm : $r = 0,97$, $P = 0,05$; pénétrateur de 1,3 mm : $r = 0,98$, $P = 0,04$). Une diminution liée à l'âge de 38 % de la rigidité de la peau a été observée chez des volontaires en bonne santé ($P < 0,05$). Les coefficients de variation pour les pénétrateurs de 0,6 et 1,3 mm étaient, respectivement, de 7,4 % et de 8,5 %. Aucune tendance liée à l'effet d'hystérésis n'a été observée dans les mesures répétées.

CONCLUSIONS: La technique d'indentation présentée était précise par rapport au dispositif d'essai des matériaux de laboratoire. En outre, les variations cutanées liées au vieillissement ont pu être détectées à l'aide de la technique d'indentation. Le nouveau dispositif s'est avéré utile pour la surveillance de la rigidité de la peau dans des conditions cosmétiques et cliniques.

Introduction

As most functions of skin depend on its mechanical properties, an understanding of tissue mechanics is important to diverse areas including medicine and the cosmetic industry [1,2]. In clinical environments, the integrity of soft tissues is often evaluated with visual inspection and manual palpation. However, this procedure is highly subjective, and dependent on the experience of the person conducting the skin assessment [3–5]. The quantitative measurement of skin elasticity may offer sensitive information on the changes of skin mechanical properties not easily detected with subjective assessment of skin appearance. In the cosmetic industry, the measurement techniques are commonly based on applications of indentation, suction, torsion or tension measurements [3,6–10] with a large number of different quantitative parameters reported. However, the physical basis and, thus, the relation of the determined parameters to skin physiology are not always clear [3]. To best interpret detected changes in skin mechanics, it is important that physiological and anatomical characteristics affecting the measurements are known. As skin has a layered structure, determination of tissue components affecting measured mechanical properties is difficult but essential for the proper design of quantitative tools used to assess skin mechanical properties. These layers, that consist of a porous solid matrix and fluid, result in complex, anisotropic, heterogeneous and viscoelastic properties [8,11]; although, the skin mechanical properties are mainly determined by collagen and elastin fibre networks along with ground substance of the dermis [8,12]. The collagen fibre network structure determines anisotropic

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and nonlinear skin properties [11,13] and permits high strains with low forces as well as high tensile strength with high forces [14,15]. Elastin is responsible for skin's mechanical integrity at low loads [15]. The ground substance, tissue water and collagen, determines skin's viscoelastic properties [11,15]. Furthermore, hydration of superficial skin has been reported to affect the mechanical properties of the skin [16]. Skin is in tension in its relaxed state, and therefore, determining its mechanical behaviour *in vivo* is complex [17].

Various indentation-based devices have been used to determine skin stiffness [1,7,18–20]. The basic technique utilizes skin compression with an indenter by measuring the force resisting the strain. Accordingly, an indentation principle is a feasible method to assess skin stiffness [21]. However, portable and easy-to-use devices enabling a fast, straightforward and reliable determination of skin stiffness, optimal for clinical and cosmetic applications, are not widely available. For robust determination of skin stiffness, uncertainty generated by layered structure of the skin on the measurements needs to be minimized. In a recent study, a finite element modelling was used to optimize the indentation geometry and to determine the tissue components affecting the indentation measurement [21]. Mechanical properties and volume of the skin were found to be dominant in determining the indentation response of forearm skin when a short indenter was used (indenter length = 0.5 mm and diameter = 2 mm) [21]. As the effect of adipose tissue and muscle on indentation response was small, it was concluded that a narrow, short and round ended indenter combined with a short measurement time might be feasible for robust and straightforward clinical determination of skin mechanical integrity [21]. As the performance characteristics of designed indentation device are still unknown, the aim of this study was to introduce a new indentation-based device, compare the technique with an industrial material tester and evaluate the fast indentation technique for the determination of age-related skin stiffness changes in healthy female volunteers.

Methods

Measurement device

Skin stiffness was determined using a commercially available hand-held device applying an indentation principle (Elastimeter, Delfin Technologies, Kuopio, Finland). Two identical devices with cylindrical indenters with diameter of 2.5 mm and lengths of 0.6 mm or 1.3 mm were used. Each device has two force sensors, one sensor is connected to the indenter and the other to a reference 23-mm-diameter base plate as shown in Fig. 1. The force required to

indent the skin to depths of 0.6 mm and 1.3 mm is determined by characteristic indenter-to-reference plate force sensor curves.

The principle of the indentation technique of this device is presented in previous papers [7,18,21]. Briefly, the device consists of two coaxial 1500 g load cells (Honeywell micro switch force sensor FSG-15N1A, Columbus, Ohio, USA). The indentation measurements are conducted by manually pressing the indenter onto skin surface. To make standardized measurements, the device monitors the speed and force applied on the skin and rejects the measurements taken, *that is* too slowly/quickly or with too low/high force. After five sequential measurements, the device calculates an average and displays the result in units of (N m⁻¹). The time to make five successful measurements is about 5–10 s.

Reference materials for assessing indenter accuracy

To evaluate indenter accuracy, the mechanical properties of five different plastic foams of varying stiffness were determined. These materials were cut into cylinders with a diameter of 19.0 ± 0.1 mm [mean \pm SD] and a height of 10.0 ± 0.8 mm. The reference material properties were determined using a custom-designed linear servo motorized material testing device (Fig. 2A) consisting of a precision motion controller (Newport PM500-C, Irvine, CA, USA), an actuator (displacement resolution of 1 μ m, Newport, PM1A11939) and a 1 kg load cell (AL311AR, Honeywell, Columbus, OH, USA).

The procedure was a one-step unconfined compression test. First, the surface of reference material was adjusted parallel with the compressing plate using goniometers (Edmund Optics Inc., Barrington, NJ, USA). Then, the compressing plate was driven into contact with the surface of reference material, and the thickness of the sample was recorded from the actuator. To ensure good initial contact between the reference material and compressing plate, the reference material was compressed to 2% pre-strain (*i.e.* 2% compression relative to sample thickness). The pre-strain level was selected to be high enough to ensure good initial contact but low enough to assure reasonable relaxation time after the pre-strain step. After a 1-h relaxation, the reference material was compressed to a 10% strain in 1 s (*i.e.* 10% compression from remaining thickness of the sample). The corresponding load was recorded via the load cell to achieve force – deformation data from which a stress – strain curve for the reference material was created (Fig. 2B and C). Elastic modulus (E) for each foam material was determined as a slope of the linear part of the stress–strain curve (Fig. 2C). Each reference material was obtained from NMC Cellfoam PLC (Laitila, Finland) with the following designations: *E30*, *L30 PS*, *PU+E25*, *S60* and *Supersael W*.

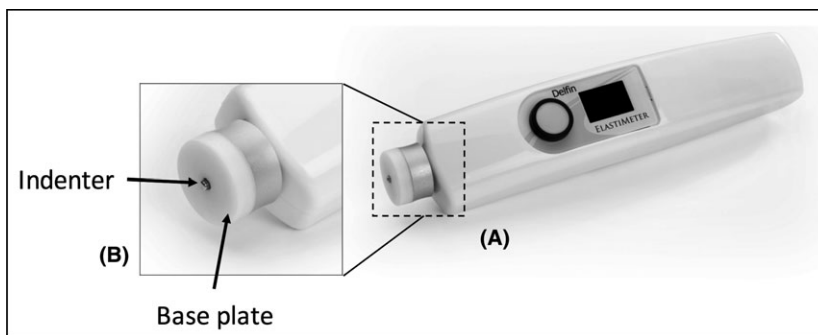


Figure 1 (A) Hand-held Elastimeter. (B) Probe of the Elastimeter with an indenter localized in the centre of the reference base plate. Essential feature of the device is a mechanical isolation of indenter from reference base plate.

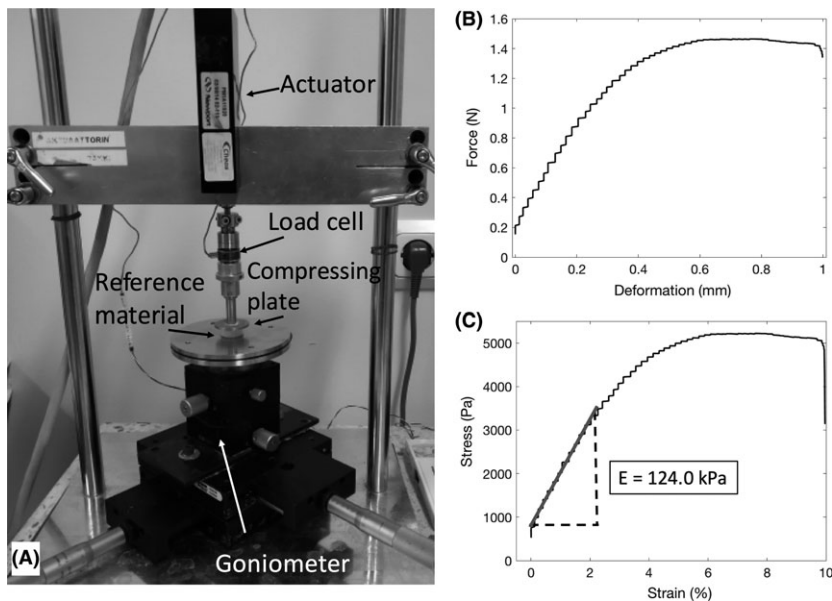


Figure 2 Test set-up and procedure for determining elastic modulus. (A) A custom-designed linear servo motorized material testing device was used to characterize the mechanical properties of reference materials. The material testing device consisted of actuator, load cell, compression plate and goniometer. (B) A representative measured compression force as a function of reference material deformation. (C) A representative stress–strain curve determined for a reference material. The elastic modulus (E) was determined in the linear part of stress–strain curve.

Indenter measurements on test materials

Each reference material type was used as a target material for the Elastimeter indentation device in which forces associated with two different indenter lengths (0.6 and 1.3 mm) were determined. The same five reference materials, for which properties were now known, were used. For the indenter tests, square planes were used with surface areas as supplied by the company of 260–650 cm² and thicknesses of 9.0–29.8 mm. For each material lying on a hard table surface, 20 repeated indentation measurements were performed. Each indentation was conducted at different locations of the material to ensure that measurements were being taken on fully relaxed materials. From the measured indenter force, a stiffness value in a unit of Newtons per metre (N m⁻¹) was calculated by dividing the measured indenter force by material deformation (length of the indenter). This stiffness unit, when applied to skin, is also referred to as Instant Skin Elasticity (ISE). The term ISE is used to emphasize that the indentation measurements are taken nearly instantaneously (within 0.5 s). Thus, for the indenter lengths used, the present measurements describe elastic, not viscoelastic properties of the skin. The stiffness values measured with the Elastimeter device were compared to the elastic modulus of the test materials as determined with material testing device via a correlation analysis.

Subjects for skin stiffness measurements

A total of 54 healthy volunteers were recruited for this study. All were from the Degree Programme in Beauty and Cosmetics, Laurea University of Applied Sciences, Vantaa, Finland. Recruiting was performed by the students under the supervision and guidance of their teachers. The selected participants gave their written consent for the measurements of the study.

Skin stiffness measurements

To evaluate various aspects of the indentation device, the study was divided into two parts.

Part 1

The capability of the device to detect age-related changes on anterior forearm skin stiffness was determined in a group of 46 seated women (age 18–79 years) with no self-reported skin diseases or medication for skin disease. Before the measurements, the volunteers were acclimatized approximately 15 min in standardized conditions (temperature: 20–22°C and relative humidity: 40–60%). Using the 1.3 mm indenter, the indentation force at the right anterior forearm was measured in triplicate at an arm site 8 cm distal to antecubital crease. Repeated measurements were conducted as close as possible at the same site. Skin stiffness was calculated as the average of the triplicate values. Subsequently, subjects were divided into six age groups (<20, 20–29, 30–39, 40–49, 50–59, >60 years, consisting of 5, 12, 7, 7, 10 and 5 subjects, respectively), and mean and standard deviation were determined for each group.

Part 2

Indentation device short-term reproducibility was evaluated by one operator. For this part, six healthy male and female volunteers (age 30–65 years) were recruited. Skin stiffness was determined bilaterally on anterior forearm sites using indenter lengths of 0.6 and 1.3 mm. At each site, measurements were repeated 10 times within a time interval of about 90 seconds. Repeated measurements were conducted as close as possible at the same site.

Data-analysis

Data-analysis was performed with MATLAB (version 7.12.0, MathWorks Inc., Natick, MA, USA). Statistical analyses were performed with SPSS (version 19, IBM, USA). Assessments of correlations between elastic moduli and stiffness determined using the plastic reference materials were made using Pearson correlations. Statistical significance in stiffness values determined among different age groups was determined with a Kruskal–Wallis one-way ANOVA. Pairwise analysis was conducted using Dunn–Bonferroni method.

Table I Elastic modulus (kPa) and ISE (N m^{-1}) values determined for the plastic foam materials ($n = 5$).

Plastic foam	Newport material testing device (kPa)	0.6 mm indenter (N m^{-1})	1.3 mm indenter (N m^{-1})
PU + E25	57.6	60	80
E30	48.2	75	64
L30 PS	131.8	88	88
S60	124.0	90	88
Superseal W	274.1	140	144
Correlation between ISE and Elastic modulus		$r = 0.97, p = 0.05$	$r = 0.98, p = 0.04$

Furthermore, the subjects younger and older than 30 years were pooled and the statistical significance in ISE values between these two age groups was determined using Mann–Whitney *U*-test. The level of significance was set to 0.05. The reproducibility of the indentation measurement was determined as a coefficient of variation (CV) [22]. The confidence intervals were calculated using non-parametric bootstrap sampling method [23].

Results

Test material elastic modulus vs. elastimeter determined stiffness (ISE value)

The elastic modulus of the plastic foam materials determined from the slope of the stress–strain curves ranged from 48.2 kPa for the E30 material to 274 kPa for the Superseal W material (Table I). The stiffness (ISE) values, determined using the 0.6 and 1.3 mm indenters, varied between 60–140 N m^{-1} and 64–144 N m^{-1} , respectively (Table I). Furthermore, the high correlation between the ISE values determined with 0.6 mm or 1.3 mm indenter and the measured elastic modulus was observed (Table I).

Age dependence of anterior forearm skin stiffness

Indentation forces, measured with the 1.3 mm indenter at forearm, ranged from 15 to 118 mN with corresponding ISE values ranging

from 12 to 91 N m^{-1} over the entire age range. A significant decrease in ISE was observed with increasing age until the 40–49 years was reached (Fig. 3). Above this age, there were slight variations in the magnitude of ISE with the appearance of a decline in ISE after age of 50–59 years. However, no statistically significant differences were found between age group above 40–49 and the ISE values of the 40–49 age group. However, when only two age groupings were considered, with group young being subjects <30 years of age ($n = 17$) and group older being subjects ≥ 30 years ($n = 29$), a highly significant difference in ISE values was found. For this analysis, ISE values for group older were significantly less than for group young ($50.4 \pm 14.4 \text{ N m}^{-1}$ vs. $66.2 \pm 10.7 \text{ N m}^{-1}$, $P < 0.001$).

Forearm skin ISE value reproducibility

The CV of ISE values determined in the six healthy volunteers was (mean and 95% confidence interval): 7.4% (4.5–8.6) and 8.5% (6.1–8.7) for indenter lengths of 0.6 and 1.3 mm, respectively. ISE values obtained using indenter lengths of 0.6 and 1.3 mm were highly correlated ($r = 0.94$, $P < 0.001$). No trend related to the hysteresis of skin was observed in the repeated measurements (Fig. 4).

Discussion

In the present study, the feasibility of a novel hand-held indentation device for determination of skin stiffness was evaluated. The stiffness values determined for reference materials with different mechanical properties correlated well with laboratory measurements taken with material testing device. Furthermore, changes in skin stiffness related to ageing could be sensitively demonstrated with the indentation device.

In the literature, several techniques for *in vivo* determination of mechanical properties of the skin have been reported [3,6–10]. The most commonly used technique to measure mechanical properties of skin is based on the measurement of skin deformation when a negative pressure is applied to the skin [24, 25]. Several parameters related to skin elasticity and viscoelasticity have been derived based on the suction measurement. However, in suction stress, the effect of skin is hard to isolate from that of adipose tissues [26]. In

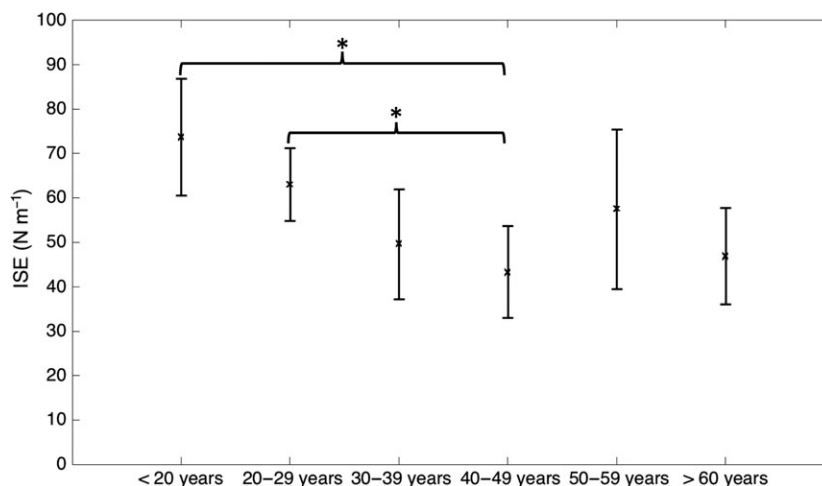


Figure 3 Mean and standard deviation of the ISE with different age groups. A decreasing trend was detected in the values of ISE as a function of age. The ISE values were increased temporarily in the age group of 50–59 years. * $P < 0.05$ Kruskal–Wallis test. The adjusted *P*-values were determined to compensate the bias caused by multiple testing.

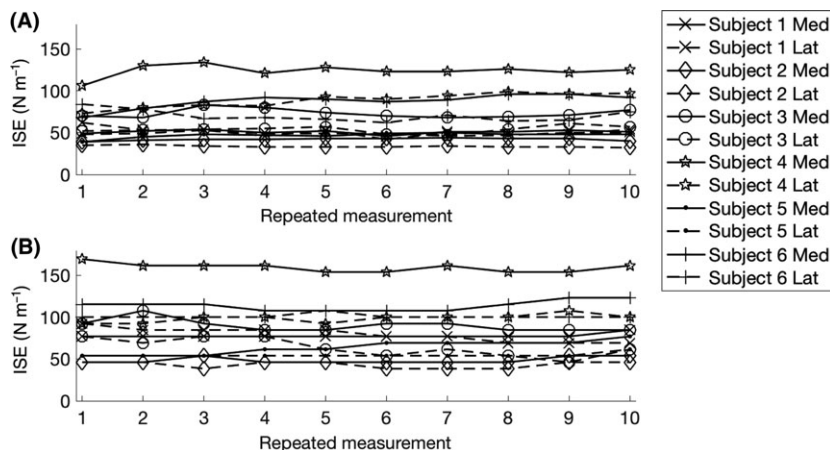


Figure 4 Short-term reproducibility measurements for (A) 0.6 mm indenter and (B) 1.3 mm indenter. Measurements were conducted on medial and lateral forearms of subjects' ($n = 6$). No trend related to hysteresis was detected from the repeated measurements.

addition, a hysteresis effect can be seen in the negative pressure suction devices since after each repeated cycle it takes longer for the skin to retract to its original position [24,25]. In the present indentation measurement, the effect of hysteresis on the repeated measurements is minor.

To determine meaningful parameters related to skin mechanics, the contributions of different skin layers on measurement techniques need to be known. Due to the complex layered composite structure of the skin, the determination of the contributions of the different tissues layers to the measured mechanical response is challenging. In previous studies, the finite element modelling was used to optimize the indentation geometry and to determine the contributions of major tissue components to indentation response of skin [21]. Skin was found to be mainly responsible for the indentation response when a short indenter (0.5 mm) was used. Furthermore, the effect of skin thickness or adipose tissues on stiffness values was found to decrease as the length of indenter was decreased from 2 to 0.5 mm [21]; thus, using a short indenter is theoretically optimal for the determination of skin stiffness. However, application of a very short indenter of 0.2 or 0.3 mm [1,27] might make the measurement prone to errors and, thus, jeopardize the reproducibility of the technique. Therefore, longer indenter, such as 0.6 mm or 1.3 mm, might be the optimal compromise for accurate and reproducible determination of skin stiffness. In the present study, the reproducibility determined with 0.6 mm was comparable to that of the 1.3 mm indenter.

High correlations were detected between the ISE values and Young's elastic moduli determined with 0.6 and 1.3 mm indenters for reference materials. The elastic reference materials were selected to cover the expected range of skin stiffness values in both healthy and pathological skin. The ISE values determined from healthy volunteers in the present study ranged between 36 and 162 N m^{-1} . The corresponding stiffness values determined for reference materials ranged between 60 and 144 N m^{-1} . However, as the majority of the measured ISE values were less than 100 N m^{-1} , the correlation analysis was performed also for reference materials with stiffness values most typical for forearm skin. Reducing the number of reference materials in correlation analysis to stiffness between 60 and 90 N m^{-1} slightly decreased the correlation between the ISE and elastic modulus ($r = 0.85$ and $r = 0.86$ for 0.6 and 1.3 mm indenters, respectively). However, after reduction of reference materials from five to four, the correlation was

not statistically significant. To achieve better statistical power in the correlation analysis, more reference materials with lower stiffness values should have been included. However, additional materials corresponding the stiffness range of the skin were not available in the present study.

Measurements in human skin yielded significant correlations between indenter lengths. The stiffness values determined in the present study with 1.3 mm indenter for the forearm were in the same range as those reported previously for a 0.3 mm indenter [27]. Mayrovitz *et al.* [27] also noticed that in facial skin the 0.3 mm indenter data correlated well with the 1.3 mm indenter results. However, the 1.3 mm indenter might be too long to assess elasticity of a thin skin, but for the determination of subcutaneous firmness, a longer indenter of 1.3 mm is feasible. Therefore, we consider the 0.6 mm indenter to be adequate for the assessment of elasticity in human skin.

The skin stiffness of healthy female volunteers was found to decrease with ageing. This finding is in line with several previous studies conducted with indentation principle [1], Cutometer [28,29] and various other techniques [27,30,31]. The age-related decrease of skin stiffness is caused by the degeneration of the skin elastin and collagen network and decrease of skin thickness [32]. The dimensions of indenters applied in the present study were designed to minimize the effect of skin thickness and the mechanical properties of adipose tissues on the measured ISE values [21]. Furthermore, in the recent study, the effect of skin water content on indentation stiffness was reported to be small [27]. Thus, the observed decrease of skin stiffness with age is most probably caused by the degeneration of the skin collagen and elastin. Interestingly, the decreasing trend in skin stiffness was momentarily reversed in the age group of 50–59 years. As all subjects in the present study were women, this finding might be due to hormonal changes related to menopause and hormonal replacement therapy. In previous studies, menopause has been related to decrease in skin collagen, glycosaminoglycan and water content [33–35]. These changes lead to decrease in skin elasticity and strength [35]. However, the hormone replacement therapy has been reported to prevent the degeneration of the skin composition and restore some of the skin mechanical function [36]. Thus, hormone replacement therapy may explain the transient increase in skin elasticity in patients in the age group of 50–59 years. Although the ISE values of the oldest age group (>60 years) were markedly reduced compared with

age groups <20 and 20–29 years, the decrease was not statistically significant due to a limited number of volunteers in the oldest age group.

There are some limitations related to the indentation technique that need to be discussed. Firstly, the measured indentation stiffness is dependent on anatomical site and the position of joints. Thus, when the skin stiffness is evaluated with indentation technique the measurement locations and position of the joints need to be standardized. Secondly, friction between indenter and skin affects the measured indentation force and, thus, the determined ISE values. As the friction between the skin and indenter could not be measured in the present study, frictionless contact between indenter and skin was assumed. However, in the recent study, the effect of friction on the measured indentation forces was evaluated by means of finite element modelling [7]. Variation in the friction coefficient between 0.1 and 0.9 changed the mean indenter force by 0.4–2.9% as compared to simulation with frictionless contact [7]. However, as compared to the age-related changes in the measured indentation forces, the uncertainty generated by unknown friction

between indenter and skin is low. Thus, the accuracy of the present measurements is sufficient for the evaluation of skin stiffness. Finally, two indentation devices with different indenter lengths were used in the present study and, thus, some of the detected differences between the devices may arise from differences in the components and calibration of the devices – not solely from difference on indenter length.

To conclude, the ElastiMeter was found to be feasible for the determination of skin stiffness. The age-related changes in the skin stiffness were detected with the device. Furthermore, the accuracy and reproducibility of the technique were found to be feasible for cosmetic industry and clinical use. As the device is battery operated and light weighted, the clinical usability of the technique is high.

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