The aim of this study was to standardize the recording technique of the SERVO MED Ep IC to determine the rest time required for human subjects to obtain stabilized values of transepidermal water loss (TEWL). Measurements of TEWL were performed on the flexor side of the forearm in healthy volunteers. First, different sites were tested on different days to study intra- and inter-individual variations. The recordings were carried out every 5 min for 2 h. Values reached a linear state after 15 min of rest. A time-course type curve of TEWL was established. Key words: evaporimeter; water loss; barrier function; non-invasive method.

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Water loss due to evaporation in the human body is an important factor in the maintenance of a controlled internal fluid and thermal balance. Continuous water exchange through the human skin involves two components: sweat-gland activity and trans-epidermal water loss (TEWL). TEWL measurements depend on the integrity of the barrier function of the skin. The barrier anatomic support is the stratum corneum (SC) (1–4), which not only limits penetration of substances in contact with the skin surface but also prevents water loss. TEWL is a passive phenomenon that corresponds to a small amount of water crossing through the SC and is dependent on the SC water vapour pressure gradient (5). TEWL is a continuous phenomenon in comparison with sweating, which is transient.

TEWL measurements are useful to estimate SC barrier capacity: (i) in assessing baseline values and factors of variation (6–8), (ii) in predicting and evaluating irritant effects of topical products (9–12), (iii) in providing a better approach to some skin disease mechanisms such as contact and atopic dermatitis for which abnormal TEWL values have been reported (13, 14). The water content of the SC and TEWL is profoundly influenced by a variety of physiological and environmental factors (15–17). These sources of variations need to be known and taken into account to obtain reliable TEWL values. Most dermatological studies today involve the evaporimeter developed by Nilsson, because of its minimal influence on the microclimate surrounding the surface of the skin and its facility of use. “Good laboratory practices” have been reported by Pinnagoda et al. (15) for TEWL measurements with the evaporimeter in dermatological use. However, the exact duration of time for human subjects to reach an equilibrium state with the environment has been accurately defined and this parameter has been estimated empirically in various different ways (14, 18–22).

The aim of this study was to further standardize the TEWL measurement method. It was necessary to confirm the literature data concerning variations related to the cutaneous site, to assess the minimal rest time required to obtain steady readings of TEWL values and to study the reproducibility of the results.

MATERIAL AND METHODS

The evaporimeter EPIC was used (Servomed, Stockholm, Sweden). The operating principle of this instrument is described in detail by Nilsson (16, 17).

Experimental conditions

Measurements were performed between March and June on adult Caucasian subjects, aged 20–55 years, after informed consent. They had no history of dermatological disease. The volunteers were resting calmly in a chair during the measurements with their sleeves rolled up prior to the first measure. Measurement sites were on the flexor side of the forearm (right or left according to randomization). A Kraft screen was fixed on the probe to eliminate flux variations due to atmospheric movements. A protection cover (n° 2108, supplied by Servomed) was used to prevent any contact between the skin and the thermistors of the probe. Sweating was not previously inhibited with an anticholinergic substance, since sweat gland activity does not interfere with TEWL values when the ambient temperature is less than 24°C (5, 6). Temperature (°C) and relative humidity (RH) were constant in the room (°C: 21.5 ± 2°C; RH: 43% ± 5%).

In the first part of the study we evaluated TEWL according to anatomical sites. In 11 subjects at rest for 30 min (this duration was chosen arbitrarily) measurements were carried out at 3 different areas on the forearm, 3 times at each site, one immediately after the other: at 4 cm (site 1), 15 cm (site 2) and 20 cm (site 3) from the wrist.

In a second set of experiments, TEWL was evaluated according to rest time: the anatomical test site was chosen according to results obtained in the first experiment (site 2). First, measurements were recorded every 5 min for 2 h on 3 different days in 7 subjects to study the reproducibility of the results (study 1). Subsequently, measurements were recorded every 5 min for 2 h on the same day in 16 subjects (study 2).

RESULTS

TEWL according to anatomical sites

A two-way analysis of variance (ANOVA) showed significant differences for subjects and sites (p < 0.001). The 3 sites showed different values (Newman-Keuls test). The highest TEWL values were obtained at site 1 (mean ± SD: 6.2 ± 0.8 g m⁻² h⁻¹) and the lowest at site 2 (mean ± SD: 2.8 ± 0.6 g m⁻² h⁻¹). Site 3 did not present significantly different values from site 2 (mean ± SD: 3.2 ± 0.7 g m⁻² h⁻¹). Only the wrist region differed significantly from other sites (p < 0.05).

A second ANOVA (on differences in the maxima and minima values) showed no influence of subjects on variability in the results, i.e. inter-subject reproducibility was good and reproducibility was the same for the 3 sites (p = 0.05).
TEWL according to rest time

Study 1. A two-way factorial ANOVA showed an influence of “time” and “subject” factors on TEWL values (p < 0.001), and there was no time-subject interaction. Multiple comparison of average values using the Newman-Keuls test (23) allowed ranking of mean values in homogeneous groups over time. There was a significant difference between values at t = 0 (mean ± SD: 6.2 ± 1.1 g m⁻² h⁻¹) and values at t = 15 min (mean ± SD: 4.9 ± 0.5 g m⁻² h⁻¹), as well as later times (p < 0.05).

Statistical analysis did not reveal any significant difference between values at t = 15 min, t = 30 min (mean ± SD: 4.7 ± 0.7 g m⁻² h⁻¹) and t = 60 min (mean ± SD: 4.6 ± 0.9 g m⁻² h⁻¹). However, data obtained at t = 120 min (mean ± SD: 4.6 ± 0.8 g m⁻² h⁻¹) also showed a significant difference with other times (p < 0.05).

The Hartley test (23), on the range of squares for each subject at each time, showed that variability for all subjects was the same and that the reproducibility was good.

Study 2. According to the preceding results, it was possible to group results obtained from 16 subjects (Fig. 1). Using a non-parametric test, the Friedman test (23), which takes into account inter-subject variability, the study 1 results were confirmed: significantly different values between t = 0 and t = 15 min (p < 0.001) and steady-state values from time t = 15 min to t = 60 min.

DISCUSSION

In the present study, experimental TEWL values ranged from 1 to 10 g m⁻² h⁻¹ and were in close agreement with those reported in previous studies according to the measurement sites (5, 24–26).

The experimental conditions were similar to those recommended by Pinna and colleagues (15) with physical parameters (RH and T), and biological and psychological conditions strictly monitored to avoid any influence on TEWL values. Age and sex of subjects were not taken into consideration for the interpretation of results, since they have no influence on TEWL (6–15), even though recent studies have pointed out a decrease in TEWL with age but for subjects that are older (70 + 13.8 years) (27) than those in our study (32.5 + 11.3 years).

The inter-individual variations in TEWL were a normal physiological reflection, with TEWL differing according to the subjects (15–20). However, since the variability of responses was the same for each subject, results obtained with different volunteers could be grouped. Studies of different areas of the forearm showed differences between the 3 sites. The site close to the wrist showed the highest values in all cases and this was in agreement with previously reported data (15, 22, 28). The nearest elbow region showed higher values than the median site, as described by Piros et al. (22). But statistically, only the wrist region differed significantly from other sites. The sweat gland density and activity varied on the forearm, increasing towards the wrist, but this could not fully explain the higher values for site 1. Another hypothesis would be that this anatomical region is more exposed to mechanical and atmospheric influences than the others and that the SC could be more easily and regularly irritated, leading to increased TEWL values. Pinna and colleagues (15) suggested an emotional influence of the first measurement on the wrist region. But in subsequent studies, the results were the same although the order of measurements was randomized (22). These findings support the exclusion of the region close to the wrist for future investigations, as previously noted (20, 22, 29). For practical reasons, the median site should be selected for subsequent work.

The rest time before a first TEWL reading is an important parameter, since it is necessary to use a time interval within which TEWL values are stabilized. This parameter has been estimated in various ways, from 10 to 60 min (14, 18, 19–22).

TEWL showed fluctuations during the first 15 min of rest, probably because of the steady-state equilibrium with an ambient atmosphere was not yet reached. Values were stabilized after 15 min and remained constant until the end of the first hour. However, there was a decrease in TEWL at 120 min, but this does not seem to be useful from a practical point of view. It was thus possible to establish a time-course type curve of TEWL (using the mean of TEWL values at different times) (Fig. 1).

Knowing the exact minimal rest time required for reliable interpretation of results means having the advantage of shorter duration experiments. During protocols including a regular participation of volunteers, this final result shows its real utility: the needed rest time, reduced to only 15 min, would make protocols less restraining and ensure better continuous participation, without impairing the reliability of the results.

In conclusion, TEWL measurements with the evaporimeter should not include the area near the wrist when the measurements are performed on the ventral forearm and the minimal rest time for steady-state values is 15 min.

REFERENCES