ELSEVIER

Journal of Pharmaceutical and Biomedical Analysis
15 (1997) 1563–1569

JOURNAL OF
PHARMACEUTICAL
AND BIOMEDICAL
ANALYSIS

Instrumentation of a handy microscopic probe for concurrent observation and measurement of active sweat secretion, and its applications[1]

Tomoya Kamei [a], Ken Naitoh [a], Koji Nakashima [a], Toshio Ohhashi [b], Shinya Kitagawa [c], Takao Tsuda [c,*]

[a] *Department of Research and Development, Suzuken, Shimizu 1-7-11, Kita-ku, Nagoya 461, Japan*
[b] *The 1st Department of Physiology, Shinshu University, School of Medicine, Asahi 3-1-1, Matsumoto 390, Japan*
[c] *Department of Applied Chemistry, Nagoya Institute of Technology, Gokiso, Showa, Nagoya 466, Japan*

Received 10 September 1996; accepted 13 November 1996

Abstract

Instrumentation for the concurrent, dynamic monitoring of active sweat glands and perspiration volume is described. A device for the measurement of the rate of sweat secretion was installed on the head part of a microscope. The combined apparatus (microscopic probe) is handy for use and its weight is very light (ca. 300 g). The microscopic probe is easily attached to the surface of human skin. The dynamic activities of the sweat glands on the forehead and nose and under the nose were observed and measured when thermal, mental and physical stimuli were applied. The activities of individual sweat glands were asynchronous when observed in units of a few seconds or less; however, they worked synchronously in a unit period of several seconds. The latter were recorded as fine peaks by a strip chart recorder. The proposed system may be useful for the study of the sympathetic nervous system, the skin sympathetic reflex and the working of sudomotor nerves. © 1997 Elsevier Science B.V.

Keywords: Sweat glands; Microscopic observation; Absolute amount of secretion; United apparatus; Nervous system; Thermal stimulus

1. Introduction

Human sweating is generally caused by thermal and mental stimuli. The former effect is very important in keeping the temperature of the human body constant. Several disorders, such as hyperhidrosis, excess sweating, and adiapneustia, are connected with human sweating, and require treatment [1–10]. Additionally conditions such as somatoform and anxiety disorders and their treatment processes may be monitored by determining the profile of the sweating rate caused by mental impulses [10]. The measurement of the amount of sweat secretion is very important in therapy and also for the diagnosis of disorders connected with sweating.

* Corresponding author.

[1] Presented at the 7th International Symposium on Pharmaceutical and Biomedical Analysis, August, 1996, Osaka, Japan.

PII S0731-7085(96)02027-4

For measuring the total amount of sweating per unit period, several methods have been used such as: the collection of single sweat droplets from the surface of the finger, covered by mineral oil, using 'Eppendorf micro tubes' [3]; and use of an iodine-starch-paper technique [6]. For the continuous measurement of the secretion rate of sweating, the following methods have been used: continuous monitoring of galvanic skin resistance [8]; continuous hygrometry in a gas stream via a small capsule by using an electrostatic capacity type hygrosensor [11–13]; and continuous measurement of electrolyte concentration in sweat based on the variation of electric conductivity using a chamber [14].

Although there are instruments for continuous measurement of the amount of sweat secretion, the number of active glands on a skin surface have not been observed concurrently. For the direct observation of active sweat glands on human skin a microscope must be used. In a previous report the authors combined a microscope-video system with a continuous measuring system for the amount of sweating [16].

In the present paper new instrumentation is described for concurrent, dynamic monitoring of active sweat glands and perspiration volume. The previously proposed apparatus [15,16] has been modified to a type of microscopic probe, which makes it possible to observe any human skin surface. The features of the new instrumentation are described together with its applications for measuring the activity of the sweat glands on the forehead, nose, under the nose and finger ridges when thermal, mental and physical stimuli are applied.

2. Experimental

Both a microscopic head and a probe with a hygrosensor were combined to form an apparatus. A schematic diagram of a handy combined apparatus is shown in Fig. 1. The instrumentation was composed of a microscopic head installed CCD in its inner body (magnification × 100 and 1000; Model 6110, Keyence, Osaka), a detector for sweating rate utilizing an electrostatic capacity type-hygrosensor (Model Kenz-Perspiro OSS-100, Suzuken, Nagoya), [11–13] a video timer (minimum unit: 10 ms; Model VTG-33, For. A, Tokyo), a VHS video cassette recorder (HV-F92, Mitsubishi, Tokyo), and a CRT (Victor, Tokyo). The hygrosensor was installed just on the microscopic head part (outer diameter; 30 mm; total lengths, 143 and 185 mm; and head caps 27 and 21 mm in height for magnifications of 100 and 1000 times, respectively). The diameters of the round open windows in the cap, which were directly in contact with the skin, were 6 and 3 mm for magnifications of 100 and 1000 times, respectively. The absolute amount of water obtained by sweating was recorded on a strip chart recorder.

2.1. Observation of skin surface

Light was emitted from the inner section of the open window of the microscopic head, using both an optical fiber and acrylic resin layer installed inside the cap. The light was controlled by a on-off foot switch. The CCD image obtained by

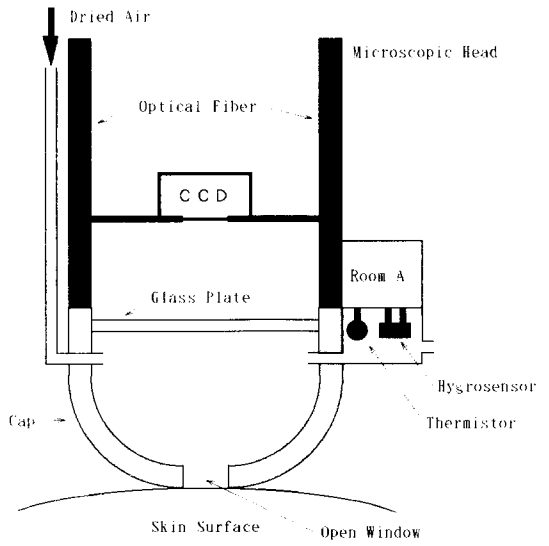

Fig. 1. Schematic diagram of a handy microscopic probe for both observation of skin surface and measurement of the rate of sweat secretion. The microscopic probe consisted of a microscopic head, its cap, hygrosensor, thermistor and an electric circuit in Room A for calculating the absolute amount of sweat rate.

the microscope was recorded on the video tape with a digital time expression. The microscopic picture could also be monitored on the CRT at the same time. The microscopic probe is so light (less than 300 g) that it is possible to place it anywhere on the human body by hand without a vibration problem.

2.2. Measurement of absolute amount of secretion

A dried air stream, 300 ml min^{-1}, obtained by passing air through a cylinder filled with silica gel (a small pump was used for the air supply), was led into the inside of the cap; then the stream was conveyed to a hygrosensor without a break in flow. The sweat secreted from the sweat glands was always diffused into the air stream and carried into the hygrosensor. The inner volumes of the caps for magnifications of 100 and 1000 were 244 and 130 μl, respectively. In an adjoining room to the hygrosensor (room A in Fig. 1), there were a capacitor with a small electric capacity, an electric circuit and a small tip for computing. The absolute amount of sweating was calculated using the following parameters: relative humidity obtained by the hygrosensor; temperature obtained by a thermistor; the capacitor as the reference for the hygrosensor; and saturated vapour pressure [11]. A skin model was used to measure both the time lag of the system and the calibration of the hygrosensor [16]. The skin model has surface regions with and without holes (the holes are similar to sweat glands); the rate of water loss from the holes can be controlled by rate of the supply of 0.9% sodium chloride aqueous solution. The latter supply was regulated by a pump (HP100-1, Denso Sangyo, Tokyo).

2.3. Thermal, mental and physical stimuli

Exercise (climbing up and down six flights of stairs, twice), hearing a loud noise from behind the head, grasping something, clenching the fist, catching a ball, taking deep breaths and mental arithmetic were used as mental impulses and physical stimuli. Five healthy men participated in two to six experiments. Of these, 2 were aged between 50 and 60, 1 between 30 and 40 and 2 between 20 and 30 years.

By attaching a strip of tape to the forehead, the area was divided into localized positions. Several holes (3 mm i.d.) were punched on the tape and numbered. The skin revealed by these holes was observed before and after exercise by using the microscopic probe.

3. Results and discussion

The handy microscopic probe is useful in observing the manner of active sweat secretion. With CRT images the direct actions of sweat secretion can be observed without any instrumental time lag. The detection system for the amount of sweat secretion should have a short instrumental time lag. After a certain period has elapsed following the application of the mental stimuli, the dynamic behaviour of the sweat glands and the response on the strip chart recorder for sweat rate can be observed concurrently. There are two other time lags caused in the human body: the period of transfer of the signal through nerve pathways in the body; and the period due to the working of sweat glands for secretion. When the instrumental time lag of the apparatus itself is very small, these periods caused in the human body system can be studied.

3.1. Measurements of time lags in the systems

These were measured as follows: first, the open window of the microscopic probe was pressed lightly on the area of the skin model without holes; and second it was shifted to the area with holes. Water loss from the holes in the skin model was detected by the hygrosensor. An accurate value for the time taken to shift the probe was recorded by using a device combining laser light and a photocell; the laser light was at first blocked by the skin model and then, when the skin model was shifted, the light was directly received by the photocell. The instrumental time lags for 100 and 1000 × magnification were 0.26 s (5 S.D. 0.018 s) ($n = 5$) and 0.24 s (S.D. 0.017 s), respectively. 10% reactive periods of the system to the level of a maximum response were 0.65 (S.D. 0.066) and 0.62 (S.D. 0.077) s for the microscopic probes of

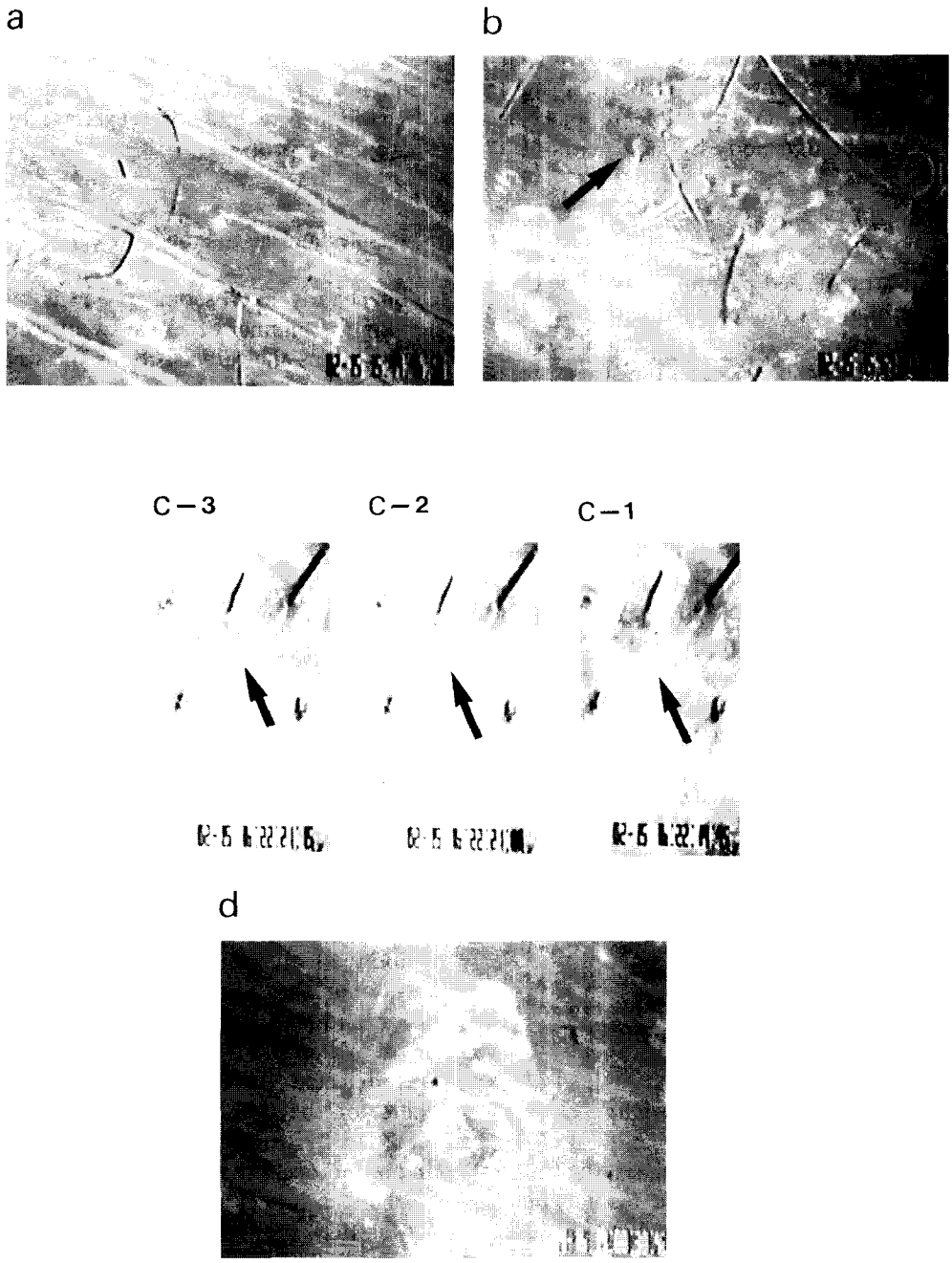

Fig. 2. Photographs of sweat secretion. (a) and (b) Secretion of sweat glands on the forehead before and after exercise (these local positions were not same); (c) a series of c-1, -2 and -3 for a single sweat gland activity under the nose was taken 0, 163/100 and 170/100 s after the impulse of a loud noise; (d) sweat secretion from a single sweat gland. Magnifications of the microscopic probe for a–c and d were 100 and 1000, respectively.

100 and 1000 × magnifications, respectively. These values of time lags are nearly same as the values given by the sweat ratementer of Kenz-perspiro-OSS-100 [13] and were small enough to use for the study of the nerve pathways and the sweat gland functions.

3.2. Dynamic observation of sweat secretion from the sweat glands

Dynamic observation of sweat secretion from the sweat glands was achieved using the present instrumentation. Typical examples are shown in Fig. 2. The sweat secretion due to thermal stress, caused by the exercise of climbing up and down flights of six stairs (temperature, ca. 25°C), was observed on the forehead before and after exercise, as shown in Fig. 2(a) and (b), respectively. Although their local positions on the forehead were different, it was evident that they became active after the exercise. The frequency of secretion of the sweat gland, shown by an arrow in Fig. 2(b), was 12–14 times per min, counted directly by using the CRT images recorded on a video tape. This frequency coincides with the frequency previously reported, that is, 5–15 [1,2]. As these values might be directly correlated with the nerve system for themoregulation, it might be possible to get information about the nerve transmission system using these observations.

Very interesting behaviour of the active sweat glands on the nose was also found. The action of sweat being forcefully expelled at intervals from the sweat glands was observed. The photographs in a series shown in Fig. 2(c)-1, -2 and -3 were taken 0, 163/100, and 170/100 s, respectively, after the impulse of hearing a loud noise. From these observations it is evident that a certain period is necessary for sweat secretion to begin after the impulse. The secretion from one sweat gland with magnification of 1000 times is also shown in Fig. 2(d). The dynamic behaviour of sweat glands on finger ridges was clearly observed with the microscopic probe. Images of such dynamic behaviour can only be obtained using the microscopic probe with high magnification.

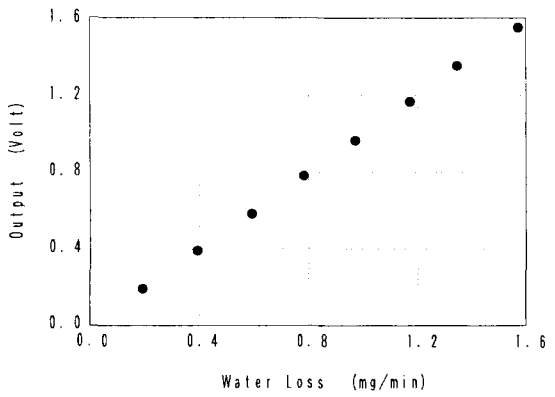

Fig. 3. Calibration curve of water loss and output signal of microscopic probe.

3.3. Calibration of sweat rate and output signal

The calibration curve of the sweating rate is shown in Fig. 3. The amount of water loss was obtained by using the skin model. It was checked experimentally that the water loss was equal to the amount of water supply to the skin model. The relationship between sweat rate (mg min^{-1}) and the amount of water loss shows a good first-order linearity. By using this calibration curve of the microscopic probe it is possible to estimate the amount of sweat secretion per unit period.

3.4. Perspiration of forehead before and after exercise

The sweating activity caused by thermal, mental and physical stimuli are demonstrated in Fig. 4. The ordinate is the sweat rate (0.1 mg min^{-1} is equal to 0.1 V output). Several interesting results are found in Fig. 4. First, the basic perspiration amount (sometimes this may include the phenomenon of 'insensitive perspiration' [1]) can be estimated from the difference between the base level of the fine peaks and the original baseline; this is specially recognized in the initial region of L_1. The basic perspiration may be the constant supply of sweat caused by thermal stimuli, and generally controlled by the central nervous system. The value of this basic perspiration after exercise, which was estimated by height of the

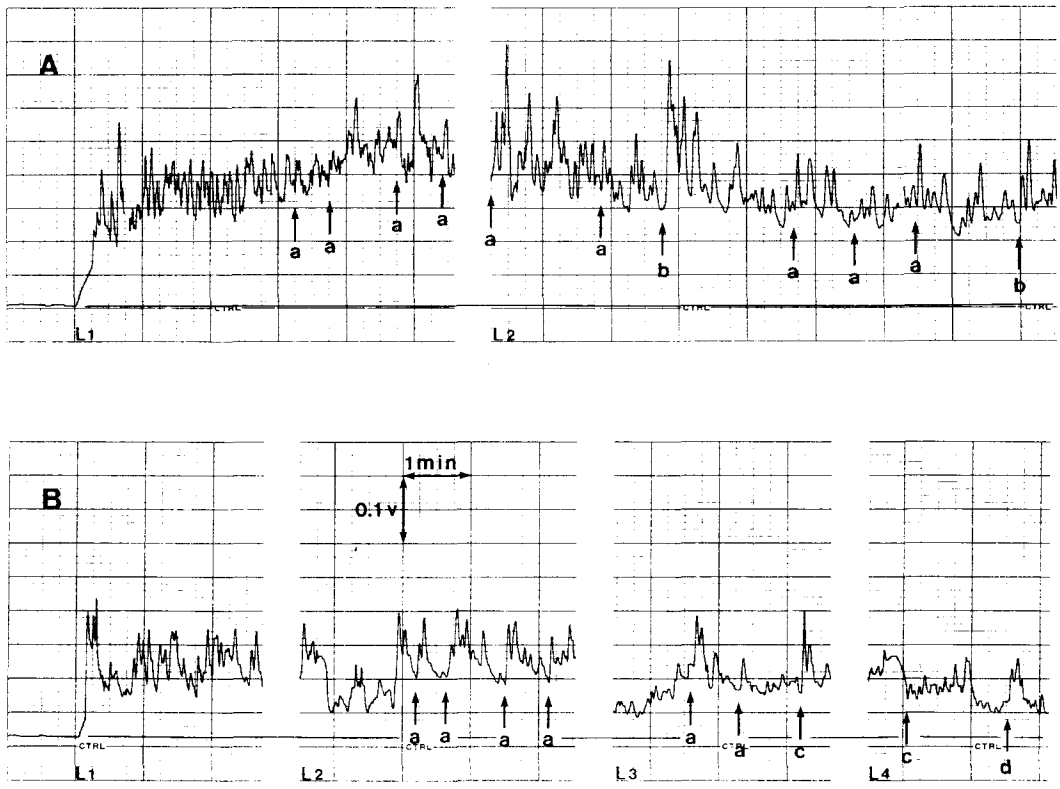

Fig. 4. Perspiration before (b) and after (a) exercise on the forehead with four kinds of impulses. Impulses (a) catching a ball; (b) hearing a loud noise; (c) mental arithmetic; and (d) clenching the fist. L_n: location of hole in the tape attached to the forehead. The suffix refers to its number. The ordinate is the output of the sweating rate, and 0.1 V is equal to 0.1 mg min^{-1}. Magnification: 100.

peak base from the original baseline, becomes double that before exercise, shown in Fig. 4. This seems reasonable because the body requires thermoregulation more after exercise than before it.

Second, the frequency of fine peaks in the region of L_1 are 14 and 10 per min for Fig. 4(a) and (b), respectively. These fine peaks reflect the activity of several sweat glands and the frequency of sweat secretion per min. Again the frequency increases after exercise.

Third, all stimuli of a–d Fig. 4(a) give high peak heights, which are estimated by subtracting the peak base from the peak maximum. The peak heights in Fig. 4(a) are almost double compared to those obtained for B in Fig. 4. These results mean that the activity of sweat glands becomes greater compared to that before exercise.

These phenomena might relate with both the central and sympathetic nervous systems.

4. Conclusions

The frequency of fine peaks per min, shown in Fig. 4, are caused by thermal sweating on the forehead. This frequency is also confirmed directly by the observation of the activities of sweat glands, which have been recorded in a video tape. The value of the frequency was very clearly confirmed.

The handy microscopic probe was used for concurrent observation and measurement of the activity of sweat secretion on the forehead, nose, under the nose and finger ridges. The apparatus is

small enough to be attached to the skin and it is possible to record the dynamic activities of sweat glands in these areas.

From the observation of live human skin under a high magnification, new information can be obtained. The proposed system can be used for the study of such things as the sympathetic nervous system; skin sympathetic reflex; the measuring of the strength of mental impulses, excitation or stress; and the working of sudomotor nerves. Attempts are now being made to apply the handy microscopic probe to skin surfaces such as axilla, forearms and legs.

References

[1] Y. Kuno, in C.C. Thomas (Ed.), Human Perspiration, Springfield, 1956.
[2] T. Ohhashi and M. Uono (Eds.), Parmer Perspiration—Measurement and Clinical Applications (in Japanese), Life Medicom, Nagoya, 1993.
[3] H.M. Emrich and H. Dalheim, Klin. Wochenschr., 55 (1977) 291–292.
[4] K.M. Main, D.A. Price, M.O. Savage and N.E. Skakkebae, J. Clin. Endocrinol. Metab., 77 (1993) 821–823.
[5] W.D. James, E.D. Schoomaker and O.G. Rodman, Arch. Dermatol., 123 (1987).
[6] J.R. Silver, W.C. Randall and L. Guttmann, J. Neurol. Neurosurg. Psychiatry., 54 (1991) 297–304.
[7] M.D. van den Broek, C.M. Bradshaw and E. Szabadi, Eur. J. Clin. Pharmacol., 26 (1984) 209–213.
[8] J.S. Vaidya and R.A. Dhuma, Ind. J. Pharmacol., 38 (1994) 319–322.
[9] S.D. Solack, G.L. Brengelmann and P.R. Freund, J. Appl. Physiol., 58 (1985) 1546–1552.
[10] T. Kato, S. Nishimiya, I. Kato and R. Inoue, Jpn. J. Persp. Res., 2 (1995) 84–86.
[11] M. Sakaguchi, N. Ono, T. Kamei, H. Yokochi, K. Nakashima and T. Ohhashi, Jpn. J. Med. Eng., 28 (1990) 137–142.
[12] Sakaguchi, N. Ono, T. Kamei, K. Nakashima and T. Ohhashi, 32nd Conf. International Union of Physiol. Sciences, Glasgow No. 175.25/P, Abstracts, August 1–6, 1993, p. 58.
[13] M. Sakaguchi, N. Ono, T. Kamei, K. Naitoh, K. Nakashima and T. Ohhashi, Jpn. J. Persp. Res., 2 (1995) 15–18.
[14] A.K.M. Shamsuddin, T. Tamura, Y. Kimura and T. Togawa, Jpn. J. Persp., Res., 2 (1995) 19–22.
[15] T. Tsuda, S. Kitagawa, T. Kamei, K. Naitoh, K. Nakashima and T. Ohhashi, 2nd Symp. Perspiration Research, Tokyo, August 19, 1994, Jpn. J. Persp. Res., (Suppl. 1) 29, 1994.
[16] T. Kamei, K. Naitoh, K. Nakashima, T. Ohhashi, S. Kitagawa and T. Tsuda, Instrumentation Sci. Technol., in press.