Original

Identification and Assessment of Carbon Monoxide in Gas Emanated from Human Skin

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Abstract

Carbon monoxide (CO) was clearly identified in gas that emanated from human skin and is called skin gas. It was collected by covering the skin surface of the subject's hand with a poly (vinyl fluoride) bag for 10 minutes and then analyzed by a GC with a semiconductor gas sensor. It was proved that CO was emanated from the human skin of six healthy volunteers. The emanated CO amounts from the hands for 10 min were 8.1 ± 4.2 ng. Furthermore, CO in skin gas correlated with that from the breath at a level of r = 0.68.

Keywords: skin gas, non−invasive, carbon monoxide

1. Introduction

Recently many people have become more health conscious and are constantly monitoring their health. Therefore, safe, easy, and non−invasive monitoring methods have been studied. Breath [1, 2], saliva, urine, and sweat [3] can be painlessly collected from humans. In addition, the gas emanating from human skin called skin gas is also collected non−invasively. Although it is difficult to determine the components emanating from human skin due to their low concentrations, we have demonstrated that hydrogen [1, 2], acetone [1, 4], methane [5], and ammonia [6] emanate from the human skin.

On the other hand, carbon monoxide (CO) in room air or the outside environment has been commonly used as an index of gas poisoning and smoking condition in humans [7]. While highly concentrated CO gas is poisonous, very low concentrations of it are useful for humans, for example, as an anti−inflammatory [8]. Furthermore, CO has drawn attention as a marker of oxidative stress [9].

The present study is the first report that clearly verifies the existence of CO in gas that emanated from human skin. Furthermore, a positive correlation was found between the CO content in skin gas and the breath of healthy humans.

2. Experimental

2.1 Materials

A poly (vinyl fluoride) (PVF) sheet and a flexible sealing film were purchased from GL Sciences (Tokyo) and the American Can Company, respectively, which were modified for the sampling of skin gas (shown in Figure 1). A standard gas mixture containing hydrogen, methane, and CO gases was supplied from Taiyo Nip-

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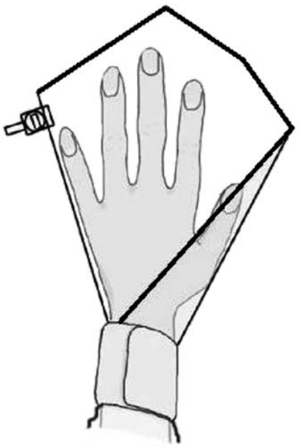

Figure 1. Skin gas sampling image

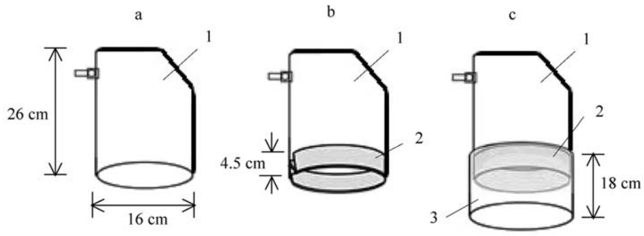

Figure 2. Modifying the bag around the wrist for skin gas sampling. 1: PVF sheet, 2: flexible sheet, and 3: polyethylene sheet.

pon Sanso (Tokyo).

2.2 Sampling bag for skin gas

Figure 2 shows the bag preparation for the sampling of skin gas. The sampling bag was made of a PVF sheet, and it resembled a glove without finger holes (Fig. 2(a)). Next, the bag's opening was covered by flexible sealing film (Fig. 2(b)). Finally, a polyethylene (PE) sheet was taped to the edge of the opening over the flexible film (Fig. 2(c)). The flexible film and the PE sheet on the sampling bag prevented skin gas leaks through the space between the bag and subjects' wrists.

2.3 Sampling procedure

Skin gas was collected by covering the left hand with a modified PVF bag (shown in Fig. 1) as follows: The left hand was washed with running tap water for 30 seconds, wiped with a paper (Kimwiper S−200, Kimberly−Clark Co., supplied by Kulesia, Tokyo), and held in room air for three minutes to dry. The hand was introduced into the modified PVF bag, which was fixed to the wrist with a band. The gas in the bag was replaced by room air and adjusted to 100 mL. While keeping the hand in the bag for ten minutes, 1 mL potion of gas in the bag was removed and subjected to gas chromatographic analysis.

Breath samples were collected as follows: Subjects held their breath for 10 seconds and about a 150 mL portion of the end−tidal air was collected in a bag made from a PVF sheet.

2.4 Subjects

Six healthy men whose ages were 39 ± 18 [mean ± SD] joined the present experiment and gave informed consent. The present study was accepted by the ethics panel of the Research Center of Health, Physical Fitness and Sports, Nagoya University (#0114).

2.5 Analytical conditions

Gas samples were analyzed by a commercially available gas chromatograph (GC) with a semiconductor gas sensor (TRIlyzer mBA−3000, Taiyo, Osaka).

Separation conditions were as follows: column temperature: 50℃; carrier gas: ultrapure air (Type: G 3, Taiyo Nippon Sanso, Tokyo, flow rate: 30 mL/min). To inject 1 mL of sample gas into this GC, hydrogen, methane, and CO gas concentrations were measured. CO retention time was 3.6 min.

2.6 Calculation of CO amount in skin gas

The amount of CO in the skin gas, *w* (ng), was calculated by the following equation:

$$w = \frac{mVM}{22.4} \qquad (1)$$

Terms *m* (ppm), *V* (mL), and *M* (g/mol) indicate the concentration of CO in the gas samples, the total gas−sample volume, and the molecular weight, respectively. In Eq. 1, 22.4 indicates the conversion factor based on the ideal gas law. $V = 100$ and $M = 28.01$ were used in Eq. 1.

In addition, the amount of CO in the skin gas samples, w_{sample}, was the sum of CO emanated from human skin $w_{skin-gas}$ and room air w_{air} because skin gas samples were collected in room air. The relationship of these values is described in following equation:

$$w_{skin-gas} = w_{sample} - w_{air} \qquad (2)$$

2.7 Statistical examination

In this study, correlation factor *r* was calculated by the linear least−squares method.

3. Results and Discussion

3.1 CO stability in the sampling bag

First, the possibility of CO loss by absorption to and permeation from the modified PVF bag was studied in the following procedure: After diluted standard CO gas was introduced to the PVF bag, 1 mL gas in the bag was successively analyzed by GC from a zero to 90 min period at about 10 min intervals. CO concentration in the bag was almost constant for 90 min at a level of 0.686 ± 0.025 ppm (relative standard deviation: RSD = 3.6%). A RSD value smaller than 5% indicates the PVF sheet can be used for CO sampling of gas that emanated from human skin.

3.2 CO identification in skin gas

Typical chromatograms of room air, skin gas, and breath sam-

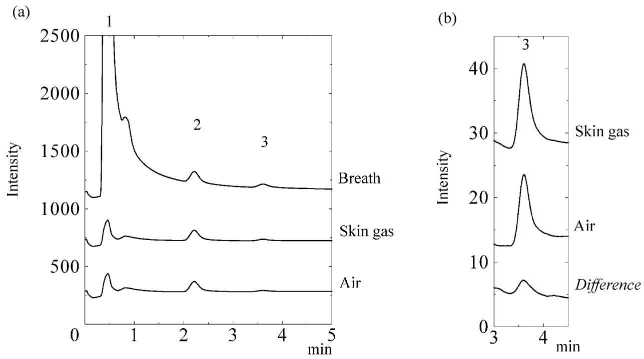

Figure 3. Typical chromatograms of room air, skin gas sample, and breath. Peak: (1) hydrogen, (2) methane, and (3) CO.

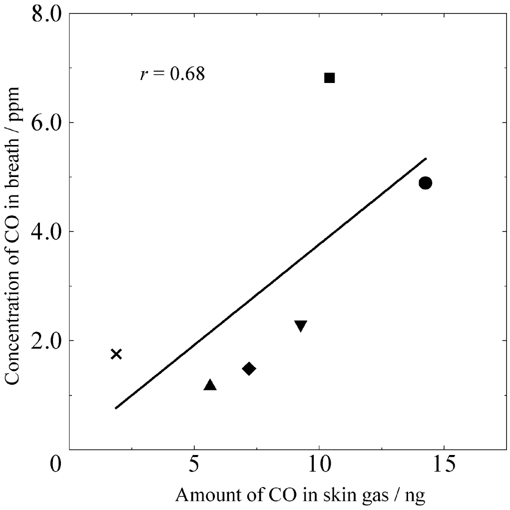

Figure 4. Relationship between CO level in skin gas and in breath.

ples are shown in Figure 3. Peaks 1, 2, and 3 were identified as hydrogen, methane, and CO, respectively. After the room air sample was analyzed as blank, skin gas and breath samples were collected and analyzed. The enlarged chromatograms are shown in Fig. 3(b). The net chromatogram of skin gas, obtained by subtracting the chromatogram of room air samples from skin gas samples, is also shown in Fig. 3(b) as *Difference*. The average amount of CO that emanated from human skin for all subjects was 8.1 ± 4.2 ng (n = 6), which was calculated by Eqs. 1 and 2. Consequently, it was clarified that CO gas emanated from human skin.

Endogenous CO is produced when heme oxygenase breaks down heme protein *in vivo* [7]. Produced CO is excreted through the lungs into the breath [10]. It is also shown that endogenous CO is excreted through human skin into the skin gas.

3.3 Correlation between CO content in skin gas and breath

The relationship between the amount of CO that emanated from human skin and breath concentration is shown in Figure 4. The result shows that CO in skin gas is correlated with breath at a level of r = 0.68.

It has been reported that CO in breath is significantly correlated with blood (CO hemoglobin) [11, 12]. Therefore, CO in skin gas may also become an indicator of CO in the blood due to the positive relationship between CO in skin gas and in the breath (Fig. 4).

4. Conclusion

To monitor endogenous CO, the measurement of CO present in blood is generally used. However, collecting blood from humans is painful. Although collecting breath is non−invasive, a mouthpiece is needed. This procedure may also encounter difficulty when used to collect from infants.

On the other hand, CO in skin gas was collected with a modified PVF bag. This method easily and painlessly collected skin gas. In addition, skin gas is stable for continuous samplings, since it emanates from the human skin all day. Thus, CO monitoring *in vivo* with skin gas can be applied to human health care in the future.

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References

[1] Naitoh, K.; Tsuda, T.; Nose, K.; Kondo, T.; Takasu, A.; Hirabayashi, T. *Instrum. Sci. Technol.* **2002**, *30 (3)*, 267−280.

[2] Kondo, T.; Tsuda, T.; Nose, K.; Ishiguro, H.; Mitsui, T.; Gao, K.; Fujiki, K. *Am. J. Gastroenterol.* **2002**, *97*, 1271−1272.

[3] Terasawa, J.; Mitsuya, K.; Ishii, A.; Tsuda, T. *Bunseki Kagaku* **2001**, *50 (12)*, 813−817.

[4] Nose, K.; Kondo, T.; Araki, S.; Tsuda, T. *Bunseki Kagaku* **2005**, *54 (2)*, 161−165.

[5] Nose, K.; Nunome, Y.; Kondo, T.; Araki, S.; Tsuda, T. *Anal. Sci.* **2005**, *21 (6)*, 625−628.

[6] Nose, K.; Mizuno, T.; Yamane, N.; Kondo, T.; Ohtani, H.; Araki, S.; Tsuda, T. *Anal. Sci.* **2005**, *21 (12)*, 1471−1474.

[7] Omaye, S. *Toxicology* **2002**, *180 (2)*, 139−150.

[8] Ryter, S.; Otterbein, L. *BioEssays* **2004**, *26 (3)*, 270−280.

[9] Montuschi, P.; Kharitonov, S.; Barnes, P. *Chest* **2001**, *120 (2)*, 496−501.

[10] Coburn, R.; Blakemore, W.; Forster, R. *J. Clin. Invest.* **1963**, *42*, 1172−1178.

[11] Wald, N.; Idle, M.; Boreham, J.; Bailey, A. *Thorax* **1981**, *36 (5)*, 366−369.

[12] Smith, C.; Guy, T.; Stiles, M.; Morton, M.; Collie, B.; Ingebrethsen, B.; Robinson, J. *Hum. Exp. Toxicol.* **1998**, *17 (1)*, 29−34.