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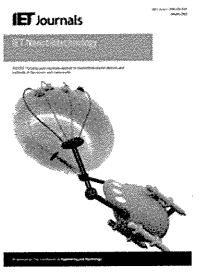

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Development of rapid oral bacteria detection apparatus based on dielectrophoretic impedance measurement method

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Abstract: In this study, a bacteria detection apparatus based on dielectrophoretic impedance measurement (DEPIM) method was demonstrated for rapid evaluation of oral hygiene. The authors integrated a micro electrode chip on which bacteria were captured by dielectrophoresis (DEP), an AC voltage source to induce DEP force, and an impedance measurement circuit to a portable instrument that enables rapid and automated oral bacterial inspection in hospitals and clinics. Special considerations have been made on effects of high electrical conductivity of oral samples on DEP force and DEPIM results. It was shown experimentally and theoretically that using a higher electric field frequency for the DEP bacteria trap and the impedance measurement could realise DEPIM application to bacteria inspection from oral samples with higher conductivity. Based on these investigations, the authors optimised the frequency condition of the DEPIM suitable for inspecting an oral sample along with the design and development of a portable DEPIM apparatus for on-site inspection of oral bacteria. Under the optimised frequency condition, DEPIM results were in good agreement with the conventional culture method showing significant applicability of the DEPIM apparatus for practical rapid oral bacteria inspection.

1 Introduction

Detection of pathogenic microorganisms is a crucial process in medical diagnosis for confirming the existence of a particular disease. Microbiological infectious disease of the oral cavity is an important matter to be concerned since the relationship between periodontal disease, caries, pneumonia, influenza and oral bacteria has been established [1–3]. Recently, aspiration pneumonia has become a focus of attention with regard to patients staying at intensive care units [4] and elderly people in nursing homes [5] because of their high mortality rate, longer length of stay in hospital and increased medical cost [6]. Yoneyama *et al.* [7] investigated the onset of pneumonia and its mortality rate among older patients in a nursing home, obtaining values of 19 and 16%, respectively, within the investigation period. Aspiration pneumonia is thought to be because of the mis-swallowing of bacteria that inhabit the oral cavity and these bacteria reaching the lower respiratory tract [8]. In addition, El-Solh *et al.* [9] suggested that dental plaque may serve as a reservoir for respiratory pathogens. Inglis *et al.* reported that the development of pneumonia depends on the amount of bacteria aspirated into the lungs from saliva being the medium that carries oral bacteria from dental plaque [10]. Influenza is a respiratory infection disease caused by the influenza viruses and has a high mortality rate in the elderly, and prevention of pandemic of influenza is a serious matter especially after a global outbreak of a new strain of H1N1 virus in 2009. Poor oral hygiene may result in increased susceptibility to influenza because bacteria enzymes may injure the oral mucosa and possibly accelerate the onset of viral infections [11]. Abe *et al.* suggest that it is necessary to accurately evaluate the amount of oral bacteria as a level of oral hygiene in order to prevent aspiration pneumonia [12] and influenza [3].

Conventionally, cultivation and colony counting techniques have been performed to evaluate oral hygiene [13] because cultivation is the most established method for inspecting the amount of bacteria not only for samples from the oral cavity but also for various samples from biogenics including humans, food, the environment etc. However, the cultivation method cannot provide a fast evaluation result since it requires rather a long time for bacteria incubation until the appearance of a visible colony on the culture medium (typically a few days); furthermore, it needs to be implemented by a specialist. Therefore in spite of this need,

evaluation of oral hygiene through the amount of oral bacteria has not been commonly used in clinical application except for research purposes. To solve these problems, several alternative bacteria counting methods have been developed. Adenosine TriPhosphate (ATP) bioluminescence is a rapid assay that detects luminescence caused by the enzyme reaction of ATP contained in various bacteria [14]. ATP bioluminescence is useful for on-site monitoring of bacterial contamination because the method does not require a culturing step, and compact equipment has been developed. However, it requires a reagent, which must be stored in a low temperature environment (typically 2 to 8°C) and must be used at room temperature. The direct-count technique using epifluorescence microscopy (EFM) is a highly sensitive bacteria detection method that requires a process of staining bacteria with fluorescent material and observation under a fluorescence microscope [15]. The measurement procedures are tedious, and inspectors need to have special skills in membrane filtration and microscopy.

Suehiro *et al.* proposed a biological cell detection technique called dielectrophoretic impedance measurement (DEPIM) based on dielectrophoresis (DEP) [16]. The DEPIM can also realise highly sensitive detection combined with electropermeabilisation [17, 18], and selective detection of biological cells according to their viability [19] or species by combining with an antigen–antibody reaction [20]. DEP is the electrokinetic motion of dielectrically polarised particles in non-uniform electric fields and is currently an active area of research in several laboratories [21, 22]. As most biological cells and macro molecules behave as dielectric particles in external AC electric fields, DEP has found many useful biotechnological applications. The DEPIM utilises positive DEP, which attracts polarised particles to a high field region, in order to capture biological cells onto an interdigitated electrode chip in the form of pearl chains. Higher cell population results in faster formation of the pearl chains, which bridge over the electrode gap and hence increase the admittance between the electrodes. By monitoring the temporal variation of the electrode impedance or admittance, the cell population can be quantitatively evaluated. By utilising positive DEP, it is possible to enrich the cell population on the microelectrode beyond that in bulk, realising highly sensitive detection of bacteria suspended in the aqueous medium. In addition, DEPIM can realise fast and simple bacteria inspection using only electrical phenomena and instruments without any preliminary chemical treatment.

The aim of this study was to adapt the DEPIM method to the detection of bacteria sampled from the oral cavity and to provide a new rapid, simple operation and on-site inspection method for the evaluation of oral hygiene through the amount of bacteria inhabiting the oral cavity. The bacterial inspection apparatus that utilises the DEPIM method should be applicable to a sample solution with high electrical conductivity. For example, a sample obtained from the oral cavity may include saliva that contains a large amount of electrolytic ions. In general, the positive DEP force becomes weak in a suspension medium with higher electrical conductivity. In order to realise DEPIM-based oral bacteria inspection, special attention was paid to the influence of suspension conductivity as well as electric field frequency on the DEP bacteria trapping process. Based on the experimental results, the DEP condition was optimised for oral bacteria detection and a hand held DEPIM apparatus was newly designed and developed, aiming at the practical application of DEPIM for the rapid and automated inspection of oral bacteria in hospitals or clinics. In addition, bacteria samples obtained from the oral cavity were inspected to validate the effectiveness of the optimised DEPIM condition and the newly developed DEPIM apparatus.

2 Material and methods

2.1 Electrode

Two different electrode configurations were used. A smooth interdigitated electrode system was employed in all the DEPIM experiments because this type of electrode configuration is suitable for accurate impedance measurement [16]. The smooth interdigitated electrode arrays of gold were patterned on a polycarbonate substrate by a laser ablation technique. Each microelectrode strip had a 5 μm gap in which cells were trapped and formed into pearl chains by positive DEP. On the other hand, a castellated electrode configuration [23] was employed for the visual observation of the cell collection process using positive DEP. The castellated electrode arrays of chrome were patterned on a glass substrate by the photolithography technique, and the microelectrode was surrounded by a silicon rubber spacer to form a chamber in which 22 μl of cell suspension liquid was stored.

2.2 DEP observation equipment

The details of the DEP observation apparatus have been described before [16]. The cell suspension liquid was stored in a reservoir tank and circularly fed to the test chamber using a peristaltic pump. Sinusoidal AC voltage was generated by a function generator (WF 1945, NF Corporation, Japan) and applied to the electrode system. Visual observation of DEP was conducted using an inverted microscope (BX-51, OLYMPUS, Japan) and a CCD digital camera (C-5060Z, OLYMPUS, Japan). The flow rate of the cell suspension liquid fed by the peristaltic pump was 2.1 ml/min, and the amplitude of the applied voltage was 10.0 V peak–peak, respectively, which were found to be appropriate conditions for the observation of positive DEP in the preliminary tests.

2.3 DEPIM equipment

Fig. 1 shows a block diagram and a photograph of the newly designed and developed DEPIM apparatus. To enable rapid and automated bacterial inspection in hospitals and clinics, the apparatus was designed as a portable instrument to enable stand-alone measurement without any other instrument or cable. The apparatus consists of two main blocks, one is the 'circuit block' which has a measurement instrument function, and the other is the 'cell block' that includes the electrode and test cell.

All the necessary functions for the electrical measurement of DEPIM are installed in the circuit block. The AC voltage source generates AC voltage, which energises the interdigitated electrode to generate positive DEP force. AC current flowing through the electrode is measured by the current detector. The current is converted from analogue to digital, and is then transferred to a digital processor. The processor calculates the electrode capacitance from the amplitudes of the applied AC voltage and detected current, and the phase difference between the two components. The sequential measurement is carried out for 20 s, and temporal variation of the electrode capacitance is stored,

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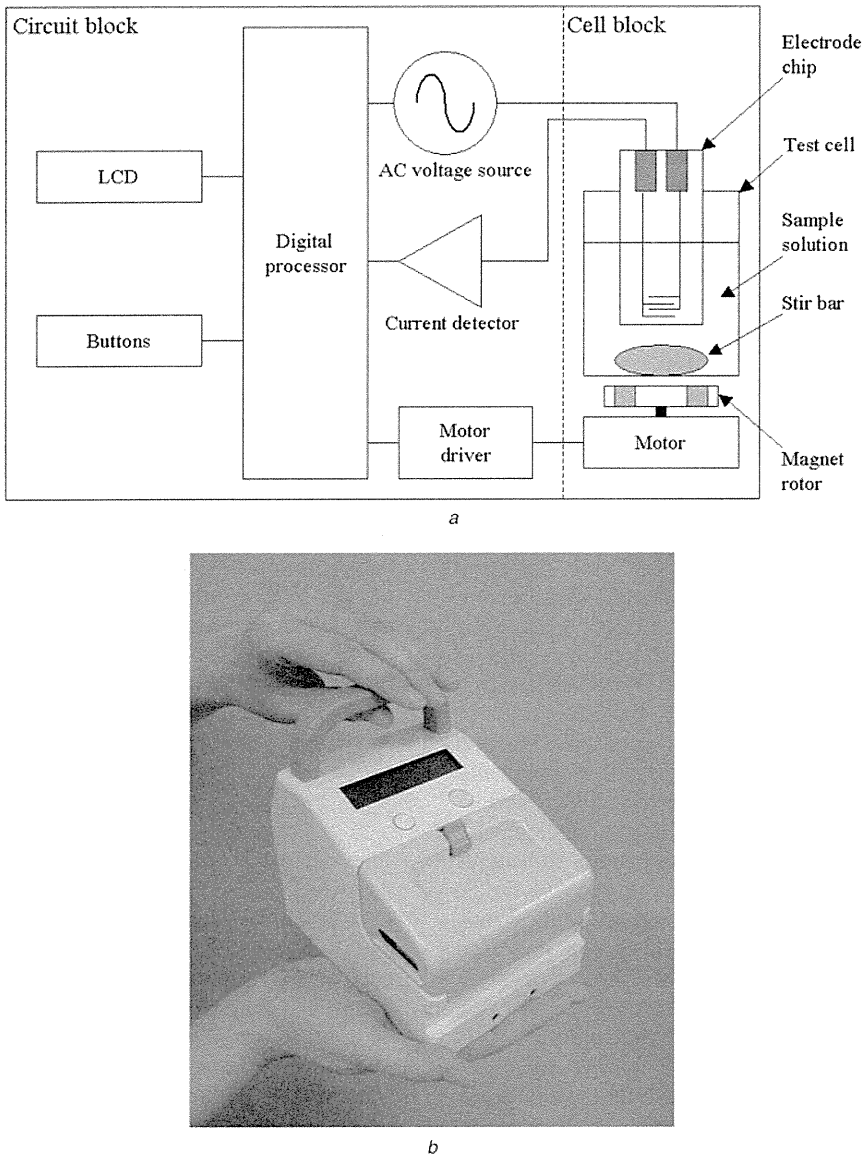

Fig. 1 *Portable DEPIM apparatus*
a Block diagram
b Photograph

then a tangent slope of capacitance change is calculated in order to estimate bacteria concentration, which has a linear relationship with the slope. According to DEPIM theory, bacteria concentration can be estimated by the increase rate of the capacitance as well as by the increase rate of the conductance [16]. In this study, the capacitance was preferred because the conductance change was expected because of the high ion concentration of oral samples and using the conductance change might have resulted in decreased accuracy regarding DEPIM. The processor also controls an LCD for the indication of the measurement results, some operation buttons, as well as a motor driver for driving a motor in the cell block to stir the sample solution.

In the cell block, 5 ml of bacterial suspension is stored in a test cell, in which the smooth interdigitated electrode is immersed. The electrode chip is connected to the AC voltage source and current detector in the circuit block. A magnetic stirrer continuously generates a circular flow in the test cell to enhance the DEP trapping of bacteria [16]. Impedance values measured by the DEPIM apparatus were calibrated using a dummy load (a parallel connection of resistance and capacitance with known values), as well as a buffer with known conductivity.

2.4 Bacteria samples

2.4.1 Optimisation of DEP condition: El-Solh *et al.* reported that *Escherichia coli* (*E. coli*) was often found in the oral cavities of elderly people and could cause pneumonia [9]. Accordingly, for observation of the DEP trapping process and preliminary optimisation of DEPIM measurement conditions, *E. coli* strain K-12 (NBRC3301), which has a high growth rate and has been successfully employed in previous work [16–20], was employed as a representative of oral bacteria in order to improve the efficiency of the experiments. *E. coli* were incubated at 30°C on agar plates for 24 h. Before each measurement, cells were harvested from the agar and suspended in a 0.1 M mannitol solution. After several washings by centrifugation, they were finally resuspended in a 0.1 M mannitol solution

(conductivity 1 μS cm^{-1}) at various diluted concentrations as determined by a colony counting method.

The oral cavity harbours a complex microbiota, with over 300 named species of bacteria so far isolated [6]. Abe *et al.* reported that bacteria exist in human saliva at a concentration ranging from 2.7×10^7 (pneumonia low-risk group) to 4.7×10^8 cm^{-3} (pneumonia high-risk group) [24]. Given that DEPIM can detect bacteria at a minimum concentration of 10^5 cm^{-3} [16], which is almost 1% of the lowest bacteria concentration expected in human saliva, the saliva can be diluted 100 times with deionised water so that the electrical conductivity is lowered to be as low as possible. According to Neyraud *et al.*, the conductivity of human saliva (mean $\pm$ SD among seven healthy subjects) were 4115 ± 1181 μS cm^{-1}, and the maximum value was 7474 μS cm^{-1} [25]. This means that the maximum conductivity of the DEPIM sample after dilution 100 times becomes about 100 μS cm^{-1}. Considering these conditions, the DEPIM experiments were conducted in the conductivity range from 5–150 μS cm^{-1}. The conductivity of each sample solution was adjusted by dissolving sodium chloride in 0.1 M mannitol solution and checked by a conductivity meter (B-173, HORIBA, Japan).

2.4.2 Verification of testing oral samples: In order to ensure applicability of DEPIM to oral bacteria detection, samples were taken from 49 elderly residents in a care facility for elderly people requiring long-term care (This study was approved by the Ethics Committee of The Nippon Dental University, School of Life Dentistry in Tokyo.). Samples were collected from the median area of the tongue in contact with the mandibular first molar using a sterilised swab (1A754S, JCB, Japan). To avoid errors caused by an inadequate sampling process, the swab was pressed against the tongue with a constant force of 0.2 N by using a special device. The swab swiped the tongue surface twice (1 cm long for each swipe). The obtained samples were suspended in 7 ml of 0.1 M mannitol, and 5 ml was used as a sample for testing with DEPIM equipment, whereas the remaining solution was employed as a sample for the conventional culture method. For the culture method, samples were appropriately diluted in 0.1 M mannitol, spread over blood agar plates (each sample, 1–2 plates) using a spiral plating apparatus (Autoplate 4000; Spiral Biotech), followed by anaerobic culture at 37°C for 48 h. These operations were executed within several hours to prevent loss of viability of the bacteria. All visible colonies grown on the plate were counted using a colony counter (Acolyte; Synbiosys), and bacterial concentrations in the samples were determined. Correlations in the data obtained by these methods were then evaluated.

3 Results

3.1 Observation of DEP trapping process of bacteria

Photographs of the DEP collection of *E. coli* are shown in Fig. 2. The DEP collection observations were made at two different electric field frequencies of 100 and 800 kHz, and two different conductivities of the suspending medium of 1 and -50 μS cm^{-1}. In the case of 1 μS cm^{-1}, bacteria were trapped around the electrode corner because of positive DEP (Figs. 2*a* and *b*) at frequencies of 100–800 kHz. On the other hand, in the case of higher conductivity of 50 μS cm^{-1}, bacteria were not trapped at 100 kHz. When forced flow was stopped, cells were focused at a weak

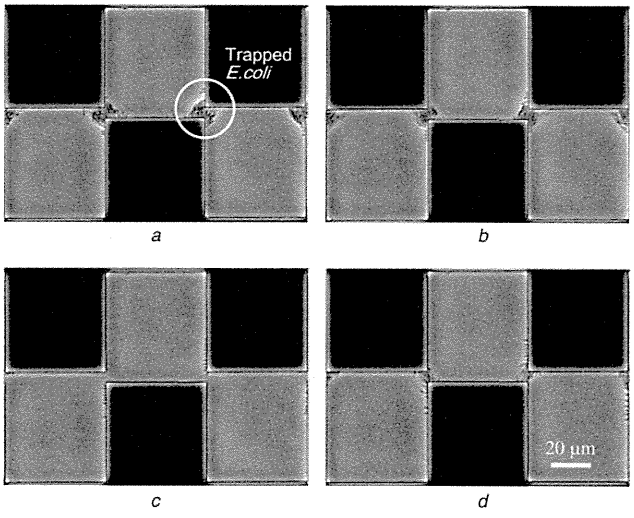

Fig. 2 *DEP collection process of E. coli*

AC signal of amplitude 10 V peak–peak was applied to a crenellated electrode for 15 s

a At 100 kHz, 1 μS cm^{-1}: bacteria were trapped by positive DEP around the electrode corner where the electric field strength becomes higher

b At 800 kHz, 1 μS cm^{-1}: the captured bacterial quantity was almost equal to *a*

c At 100 kHz, 50 μS cm^{-1}: hardly any bacteria were captured

d At 800 kHz, 50 μS cm^{-1}: the captured bacteria were slightly fewer than *a* and *b*

electric field region at the centre of the electrode gap surrounded by the electrode edge, showing that negative DEP became dominant under this condition (not shown in the picture). Some bacteria were captured at 800 kHz, although the number of trapped cells was slightly decreased compared to the case of the 1 μS cm^{-1} conductivity (Figs. 2*c* and *d*). These observation results suggest that positive DEP force exerted on the bacteria becomes weak with increased conductivity at the 100 kHz frequency.

3.2 DEPIM measurement using E. coli samples

DEPIM experiments were conducted in this range of conditions for 1–135 μS cm^{-1}. Fig. 3 shows temporal variation of the electrode capacitance increment measured with *E. coli* at 5×10^6 CFU/ml (at 100 kHz) and 2×10^7 CFU/ml (at 800 kHz). As explained in the previous work [16], the capacitance increase is because of the presence of bacteria that are trapped and enriched in the electrode gap. It was found that the capacitance increase rate at $t = 0$ was directly proportional to the cell population that was quantitatively evaluated by the DEPIM method.

At a frequency of 100 kHz, the capacitance increase rate in the case of 25 μS cm^{-1} was obviously lowered in comparison with 1 μS cm^{-1} (Fig. 3*a*). However, at a higher frequency of 800 kHz, the temporal change of capacitance was almost the same for both the conductivities of 1 and 50 μS cm^{-1} (Fig. 3*b*).

4 Discussion

The DEP force acting on a spherical particle of radius r suspended in a medium of permittivity ε_s is given by [26]

$$F_{DEP} = 2\pi r^3 \varepsilon_s \mathrm{Re}[K(\omega)] \nabla E^2 \quad (1)$$

where E is the magnitude root mean square of the applied field and Re[$K(\omega)$] is the real component of the Clausius–Mossotti

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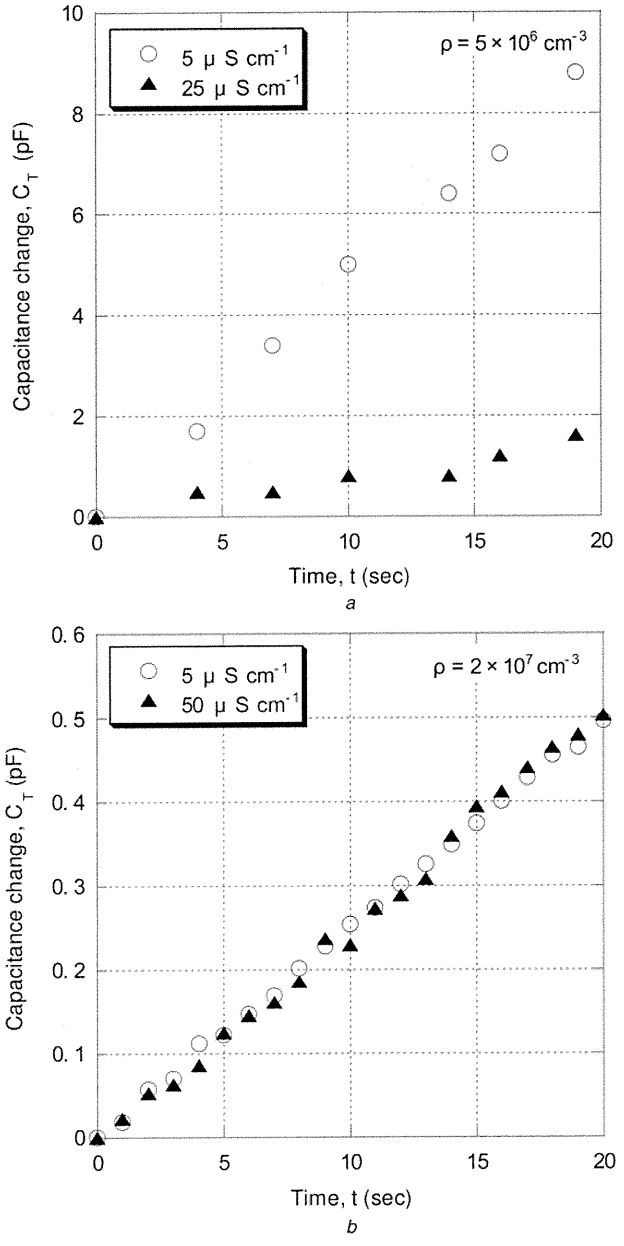

Fig. 3 *DEPIM results measured with E. coli*

a At a frequency of 100 kHz, and conductivity of 5 μS cm^{-1} (○), the capacitance increased almost linearly with time. The capacitance increase implies that bacteria were trapped and enriched under positive DEP at the electrode gap. With higher conductivity of 25 μS cm^{-1} (▲), the capacitance change became less remarkable with lower medium conductivity
b At 800 kHz, the rate of capacitance increase with time was almost the same as for the medium conductivity of 5 μS cm^{-1} (○) and 50 μS cm^{-1} (▲)
AC signal of amplitude 5 V peak–peak was applied to a smooth interdigitated electrode

factor given by

$$K(\omega) = \frac{\varepsilon_p^* - \varepsilon_s^*}{\varepsilon_p^* + 2\varepsilon_s^*} \tag{2}$$

where ε_p^* and ε_s^* are the complex permittivity of the particle and surrounding medium, respectively. For a real dielectric, the complex permittivity is defined as

$$\varepsilon^* = \varepsilon - j\frac{\sigma}{\omega} \tag{3}$$

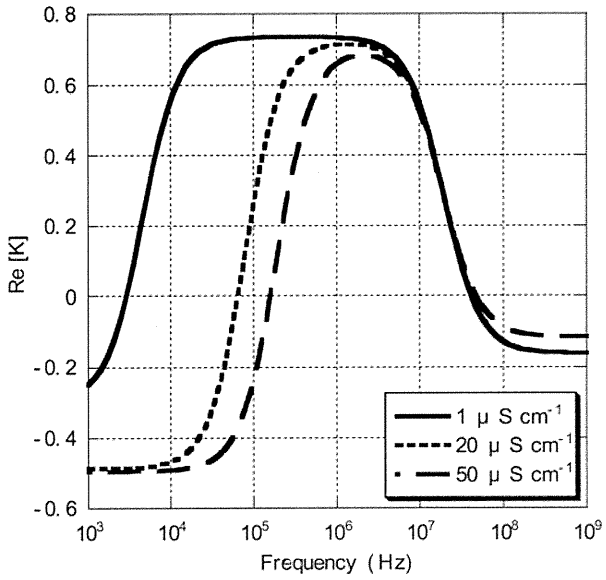

Fig. 4 *Theoretical prediction of the external medium conductivity σ_s dependency of Re[K] spectra*

E. coli cells are modelled as a dielectric sphere covered by two shells. Parameters used in calculation are listed in Table 1

where ε is the permittivity and σ is the conductivity of the dielectric and ω is the angular frequency of the applied field. The effects of the conductivity of the surrounding medium on DEP force have been attributed to the modification of parameter Re[$K(\omega)$]. In general, positive-DEP force acting on bacteria decreases with a rise in the conductivity of the surrounding medium. Muller *et al.* [27] explained that DEP force is dependent on the relative magnitude of the conductivity and permittivity of cells and that of the media. From (2) and (3), one can understand that the positive DEP force decreases together with a rise in the conductivity of the medium if the conductivity of the cell and the permittivity of the cell and medium are constant.

An example of a theoretical prediction of the suspension medium conductivity dependency of parameter Re[$K(\omega)$] is shown in Fig. 4. One *E. coli* cell is modelled as a dielectric sphere covered by shells. The shells represent the cytoplasmic membrane and the sphere covered by the shells represents the cytoplasm. The complex permittivity of the particle ε_p^* in (2) is replaced with an effective complex permittivity of *E. coli* cell ε_{eff}^*, which can be calculated by using the 'smeared-out sphere' model [28]. Parameter values of *E. coli* are determined referring to the referenced literature [29] and listed in Table 1. Fig. 4 indicates that Re[$K(\omega)$] or the DEP force decreases with increases in the medium conductivity σ_s at a lower field frequency. For example, when the medium conductivity increases from the

Table 1 Parameter values used in the theoretical prediction of Re[$K(\omega)$] shown in Figs. 4 and 5

Component	Parameter	Value
cell	radius	1 μm
cell cytoplasm	relative permittivity	60
	conductivity	0.2 Sm^{-1}
cell membrane	relative permittivity	6
	conductivity	0.25 μ Sm^{-1}
	thickness	8 nm
suspension medium	relative permittivity	80

initial value of 1 to 50 μS cm^{-1}, the Re[$K(\omega)$] value changes from a positive to a negative value, that is, DEP changes from positive-DEP to negative-DEP at the field frequency 100 kHz. This suggests that *E. coli* cells are not captured at the electrode gap by DEP under the condition of 50 μS cm^{-1}. On the other hand, the DEP force is hardly dependent on σ_s at 800 kHz. The theoretical calculations agree well with the experimental results shown in Fig. 2 where DEP collection of *E. coli* is observed only for low medium conductivity (1 μS cm^{-1}) at 100 kHz but no clear differences are observed with a rise in medium conductivity until 50 μS cm^{-1} at 800 kHz.

As explained in the previous work [19], the conductance and capacitance increase rate as a DEPIM value strongly depend on the DEP force acting on the bacteria to be detected. The weaker the positive-DEP force acting on the bacteria, the fewer the bacteria captured at the electrode gap, resulting in decreased impedance change detected by DEPIM. Fig. 5 shows effects of the suspension medium conductivity on the DEPIM measurement as well as theoretical calculations of Re[$K(\omega)$], which are normalised to the value at low medium conductivity (1 μS cm^{-1}). At a frequency of 100 kHz, the predicted positive-DEP force rapidly decreases with a rise in medium conductivity, changing to negative-DEP at 25 μS cm^{-1}. On the other hand, at 800 kHz, the decrease in positive-DEP force is predicted to be suppressed. The measured DEPIM value agrees with these theoretical predictions at both 100 and 800 kHz. The normalised DEPIM values were around zero despite the normalised Re[$K(\omega)$] value being around −4 under the condition of 50 μS cm^{-1}, 100 kHz. In the light of the DEPIM theory, it can be understood that the DEPIM value will not be less than zero even if Re[$K(\omega)$] is negative. These results indicate that frequency of 800 kHz is more appropriate than 100 kHz for DEPIM measurement of sample with high medium electrical conductivity, σ_s.

In order to ensure applicability of DEPIM to oral bacteria inspection, bacteria were taken from oral cavity of 49 elderly persons, and these samples were measured by DEPIM equipment. The conductivities of suspended samples for DEPIM measurement (mean ±SD among 49 subjects) were 51 ± 37 μS cm^{-1}, and the maximum value was 144 μS cm^{-1}. Considering the rather high electrical conductivity as well as the experimental and calculation results obtained for *E. coli* samples, measurement was carried out at frequency of 800 kHz.

Fig. 6 shows a relationship between the bacteria concentration obtained by the culture method and the tangent slope of capacitance change measured by DEPIM apparatus. As shown in Fig. 5, the DEPIM value changes with an increase in medium conductivity even if the bacteria concentration is the same. Therefore measured DEPIM values were calibrated based on the conductivity dependence shown in Fig. 5 so that effects of sample conductivity variation, which were difficult to control for the oral samples, could be cancelled out. Pearson's correlation coefficient was $R = 0.90$, and the range of error calculated from the standard curve obtained by the measurement data was −86–285%. Considering the correlation between the colony counting method and ATP bioluminescence assay ($R = 0.851$) [14] and the fluorescent counting method ($R = 0.75$) [30], correlation between DEPIM measurement and the standard culture method was significantly high.

As calculated in Fig. 4, it is predicted that stronger positive-DEP will occur in the range of MHz rather than 800 kHz under higher medium conductivity. Therefore higher field frequency will be a more suitable condition. However, a rise in the field frequency will require more careful design and probably higher cost of the impedance measurement circuit. The development of an impedance measurement circuit with a higher frequency AC source for DEP, and a current detector that can sense higher-frequency AC current accurately, will be future tasks.

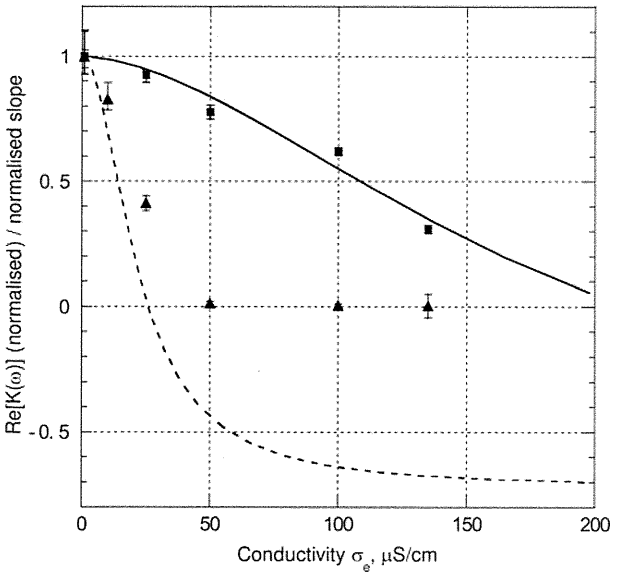

Fig. 5 *Theoretical calculations of Re[K(ω)] (solid, 800 kHz; dashed, 100 kHz) and experimental results of DEPIM at E. coli concentration of 5 × 10^5 CFU/ml (■, 800 kHz; ▲, 100 kHz), these were normalised to these values at low medium conductivity (1 μS cm^{-1})*

DEPIM measurement was carried out three times under each set of conditions. The conductivity dependencies of the Re[$K(\omega)$] and DEPIM results are almost identical, showing that the data can be employed to calibrate DEPIM results obtained for various conductivity values

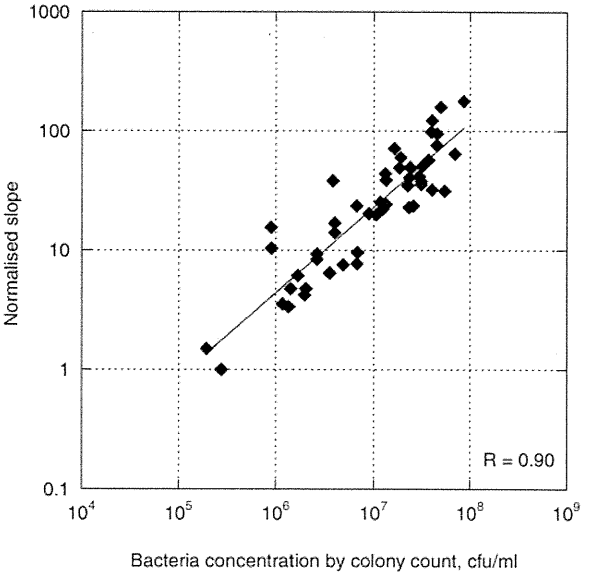

Fig. 6 *Relationship between bacteria concentration determined by conventional colony counting method and tangent slope of capacitance measured by DEPIM equipment for 49 samples obtained from the human oral cavity*

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5 Conclusions

In this article, we have described the optimisation of AC electric field frequency in the DEPIM method to enhance the measurable range of conductivity of the sample solution to adapt the DEPIM method for the inspection of bacteria obtained from the oral cavity. The optimisation was based on the theoretical calculation of DEP force dependency on the applied field frequency in the range of the assumed medium conductivity of the sample obtained from the oral cavity. Observation of DEP-trapping process of cell was carried out to find the appropriate field frequency condition in the range of the assumed medium conductivity. Based on the investigation, the performance of the developed DEPIM apparatus was confirmed by measuring *E. coli* suspended in the medium conductivity range. By applying an AC electric field of 800 kHz, the measurable range of medium conductivity using developed apparatus was expanded up to 135 $\mu S\ cm^{-1}$, at which point almost all samples from the oral cavity will be covered. Consequently, it was demonstrated that the apparatus is useful in the evaluation of the bacterial contamination of clinical samples from the oral cavity for quantitative evaluation of oral hygiene. In addition, the developed apparatus will be applied to other fields in which the investigation of the sample including ionic substances is necessary, for example, any clinical samples besides those taken from the oral cavity, as well as fields relating to the environment and the food industry.

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ORIGINAL ARTICLE: EPIDEMIOLOGY, CLINICAL PRACTICE AND HEALTH

Correlation between dental and nutritional status in community-dwelling elderly Japanese

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Aim: The purpose of this study was to clarify the correlation between dental and nutritional status among community-dwelling elderly Japanese people.

Methods: The subjects were 182 elderly individuals, aged 65–85 years, who voluntarily participated in a health seminar at Kyoto Prefectural University of Medicine. These subjects were divided into two groups according to the occlusion. The subjects in the retained contact group were those who had retained molar occlusion with natural teeth. The lost contact group were those who retained molar occlusion with removable partial dentures. Anthropometric variables such as body mass index (BMI) were collected and dietary intake was assessed using a brief self-administered diet history questionnaire (BDHQ).

Results: No statistical difference in BMI or intake of macronutrients was found between these two occlusal groups. The lost contact group reported significantly lower consumption of vegetables and higher consumption of confectionaries (foods rich in sugar) than did the retained contact group ($P < 0.05$), and therefore had significantly lower intake of vitamin C and dietary fiber ($P < 0.05$).

Conclusion: It can be concluded that natural tooth contact loss in the posterior region affect the intake of vitamins and dietary fiber. **Geriatr Gerontol Int 2011; 11: 315–319.**

Keywords: elderly, nutrition, occlusion, tooth loss, Japanese.

Introduction

The intake of nutrients as a result of food consumption is fundamental to maintaining life.[1] Many reports have shown that low food volume and an unbalanced diet are related to tooth loss, poor occlusion and other oral pathological conditions.[2–5] In particular, individuals with fewer teeth tend to avoid raw fruits and vegetables, thus reducing their intake of vitamins and dietary fiber.[6,7] Adequate intake of these nutritional elements is thought to prevent cardiovascular disease, cancer and other systemic conditions.[8,9]

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Author contribution: Mitsuyoshi Yoshida conceptualized and designed study, analyzed and interpreted data, wrote and edited the manuscript; T. K. collected data about nutritional status; M. Y. and K. T. collected data about dental examination; M. K. collected data about physical examination; and Y. A. supervised study concept and design.

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Many previous studies on this topic have been conducted in Western countries and concluded that tooth loss affects elements of nutritional intake such as dietary fiber and vitamins. However, only a few studies have been conducted previously in Japan[10–12] and may not reach a consensus. Yoshihira *et al.*[10] found no significant difference between the number of teeth present and the intake of vitamin C or dietary fiber among 57 healthy 74-year-old elderly Japanese people. However, the subjects in their study were divided into two groups only with teeth numbers as more than 20 teeth or less. We hypothesize that natural tooth contacts in the molar region are more important to the consumption of foods requiring more mastication and, as a result, this study was conducted to clarify the correlation between natural tooth contact loss and nutritional intake of vitamins and dietary fibers among community-dwelling elderly Japanese people.

Methods

The subjects were 182 healthy elderly Japanese (60 men, 122 women) aged 65–85 years living in Kyoto and participating in a health seminar sponsored by Kyoto Prefectural University of Medicine. According to brief medical interview, anyone who had a history of cardiovascular disease was excluded from the study because they had some risk completing physical assessment (original purpose in this seminar). This study was approved by the Ethics Committee at Kyoto Prefectural University of Medicine. All subjects gave verbal informed consent.

Based on oral examination, the subjects were divided into two groups according to the Eichner Classification.[13] This classification was based on existing natural tooth contacts between maxilla and mandible in the bilateral premolar and molar regions (presence of tooth contact defined as presence of a natural tooth on the maxilla and corresponding mandible, including wisdom teeth, but excluding remaining roots or root caps). Class A represents contact in all four support zones. Class B represents contact in three to one zone (B1–B3) or in the frontal region only (B4). Class C represents absence of tooth contact. The retained contact group consisted of those classified as Eichner A or B1–B3, who had retained molar occlusion in at least one molar region with natural teeth. The lost contact group consisted of individuals classed as Eichner B4 or C who had no occlusal contact with natural dentition in the molar region. All subjects in the lost contact group used removable partial dentures.

Anthropometric measurements were as follows. Body mass index (BMI, kg/m^2) is defined as the weight in kilograms divided by height in meters squared. Mid-upper arm circumference (AC, cm) was measured on the left arm with a tape measure. Triceps skinfold (TSF, cm) was measured with Harpenden calipers over the triceps muscle at the midway point between the acromion and the olecranon process. Three repeat measurements were taken to the nearest 0.5 mm, with the mean taken as the true value.

Food intake was assessed with a brief self-administered diet history questionnaire (BDHQ) along with interviews by a dietitian. The BDHQ had been developed by item-reduction from a validated self-administered diet history questionnaire (DHQ) used earlier.[14] The BDHQ is a 4-page structured questionnaire with 75 questions (55 relating to food consumption and 17 to cooking and dietary behaviors). The questionnaire assessed dietary habits during the preceding month. From this information, energy, nutrient and food intakes were calculated using an ad hoc computer algorithm for BDHQ. The validity of this questionnaire was established elsewhere that average Pearson correlation coefficients for 37 nutrients between nutrient intakes assessed with BDHQ and 16-day dietary record in adults was 0.48 in 92 men and 0.49 in 92 women.[15]

Using information from the questionnaires, nutrient intake and the intake of protein, fat and carbohydrate were calculated and evaluated in terms of the macronutrient intakes of the two groups (Table 3) based on the Standard Tables of Food Composition in Japan (5th ed.).[16] The intake of vitamins and dietary fiber were per 1000 kcal were also calculated. Multivariate ANOVA was used to compare the two occlusal groups with sex and age as covariates. Age was categorized into four groups: 65–70, 71–75, 76–80 and 81–85 years. SPSS ver. 15 was used for these analyses and all statistical significance levels set at $P < 0.05$.

Results

The retained contact group included 138 subjects (41 men, 97 women, average age 74.4 ± 3.6 years) and the lost contact group included 44 subjects (19 men, 25 women, average age 77.0 ± 5.3 years). The distributions in terms of sex did not differ, but the members of the lost contact group were significantly older than the retained contact group ($P = 0.004$). The comparisons of body composition between the two groups are shown in Table 1. Measures of AC and TSF were significantly lower in the lost contact group.

According to the data calculated from the BDHQ questionnaires, the lost contact group consumed significantly fewer vegetables and more confectionaries than the retained contact group (Table 2). Energy intake was not significantly different between the retained contact group and the lost contact group (1934 ± 59 vs 2057 ± 76 kcal/day, respectively, $P = 0.132$). There was no statistical difference between the two occlusal groups in macronutrients. Protein and carbohydrate were in the range of the Standard Tables of Food Composition in

Table 1 Comparison of anthropometric variables (adjusted mean ± standard error) for the two occlusal groups

	Retained contact group ($n = 138$)	Lost contact group ($n = 44$)	P-value
Body mass index (BMI, kg/m^2)	22.8 ± 2.8	21.9 ± 2.4	0.075
Upper arm circumference (AC, cm)	25.8 ± 2.3	24.7 ± 2.1	0.004*
Triceps skin fold (TSF, mm)	16.1 ± 5.7	12.7 ± 5.1	0.000*

P-value by multivariate ANOVA for comparison between groups adjusted for sex and age (*$P < 0.05$).

Table 2 Selected food group intakes (g/1000 kcal, adjusted mean ± standard error) for the two occlusal groups

Food group	Retained contact group	Lost contact group	P-value
Meat	34.1 ± 2.9	30.4 ± 3.7	0.507
Fish	64.5 ± 4.6	64.1 ± 5.9	0.632
Egg	19.6 ± 1.8	19.7 ± 2.3	0.821
Soy products	43.6 ± 3.0	35.4 ± 3.8	0.162
Vegetables	179.4 ± 9.9	144.4 ± 12.8	0.048*
Fruits	68.5 ± 6.0	52.6 ± 7.8	0.096
Cereals	136.5 ± 8.8	147.9 ± 11.3	0.561
Confectioneries	22.8 ± 2.7	35.8 ± 3.4	0.005*

P-value by multivariate ANOVA for comparison between groups adjusted for sex and age (*$P < 0.05$).

Table 3 Macronutrient intakes (adjusted mean ± standard error) for the two occlusal groups

Variable	Retained contact group	Lost contact group	P-value	Standard Tables of Food Composition in Japan (5th ed.)
Protein (% of energy)	17.4 ± 0.5	17.0 ± 0.6	0.317	<25
Fat (% of energy)	25.8 ± 0.6	25.9 ± 0.8	0.971	15–25
Carbohydrate (% of energy)	53.5 ± 0.9	54.4 ± 1.2	0.461	50–70
Dietary fiber (g/1000 kcal)	8.49 ± 0.28	7.36 ± 0.37	0.036*	10

P-value by multivariate ANOVA for comparison between groups adjusted for sex and age (*$P < 0.05$).

Japan, whereas fat was slightly over the standard range in both groups. Dietary fiber consumed by the lost contact group was significantly lower than for the retained contact group and, furthermore, intake of dietary fiber for both groups was lower than the recommended amount (Table 3). The lost contact group also consumed significantly less vitamins including carotene, vitamin K, vitamin B_1, vitamin B_6 and vitamin C (Table 4).

Discussion

The results from this study suggest that occlusal status based on natural tooth loss is associated with nutritional intake in dietary fiber and vitamins from food in Japan.

Chewing efficiency, for example, the rate of breakdown of food during mastication, is clearly correlated with features of the dentition such as number of posterior teeth and occlusal relationships.[17] As chewing efficiency declines, individuals report increasing difficulty chewing and may avoid difficult-to-chew foods and prefer soft and easy-to-chew foods.[18] The most pronounced difference in intake involves hard-to-chew foods such as vegetables and some fruits and a higher consumption of confectionaries, which are likely to be most affected by tooth loss.[19] The findings of this study are almost the same results as those of previous studies, and therefore may support a reasonable consensus that tooth loss affects elements of nutritional intake such as dietary fiber and vitamins in Japan.

Because all subjects in the lost contact group in our study had removable dentures, it was impossible to compare the difference in nutritional intake with and without dentures. Many previous studies have reported no significant difference in nutrient intake between patients retaining their own teeth and those with dentures, however.[20,21] Wöstmann *et al.*[22] reported that despite the highly significant improvement in masticatory ability after the optimization of dentures,

Table 4 Vitamin intakes (adjusted mean ± standard error) for the two occlusal groups

Variable	Retained contact group	Lost contact group	*P*-value
Retinol (μg/1000 kcal)	412.0 ± 85.6	496.9 ± 110.3	0.455
Carotene (μg/1000 kcal)	2475 ± 150	1790 ± 193	0.014*
Vitamin D (μg/1000 kcal)	11.8 ± 0.9	12.1 ± 1.2	0.882
Vitamin E (mg/1000 kcal)	4.42 ± 0.13	4.09 ± 0.17	0.193
Vitamin K (mg/1000 kcal)	242.0 ± 12.1	200.8 ± 15.6	0.039*
Vitamin B_1 (mg/1000 kcal)	0.48 ± 0.01	0.43 ± 0.02	0.029*
Vitamin B_2 (mg/1000 kcal)	0.87 ± 0.03	0.83 ± 0.04	0.141
Niacin (mg/1000 kcal)	10.9 ± 0.4	10.3 ± 0.5	0.182
Vitamin B_6 (mg/1000 kcal)	0.83 ± 0.02	0.74 ± 0.03	0.009*
Vitamin B_{12} (μg/1000 kcal)	7.44 ± 0.51	7.59 ± 0.66	0.750
Folate (μg/1000 kcal)	239.1 ± 9.8	212.0 ± 12.6	0.104
Pantothenic acid (mg/1000 kcal)	4.12 ± 0.11	3.93 ± 0.14	0.146
Vitamin C (mg/1000 kcal)	75.7 ± 3.3	60.2 ± 4.3	0.013*

P-value obtained by multivariate ANOVA for comparison between groups adjusted for sex and age (*$P < 0.05$).

no general improvement in nutritional status was observed. Our finding supports these previous studies that the lost contact group was associated with the lower amount of hard-to-chew foods such as vegetables that individuals consume even though they used their dentures during food intake. These results may indicate that denture use is not enough to compensate for natural teeth. Recently, Bradbury *et al.*[23] demonstrated that food instruction encourages an increase in the consumption of vitamins and minerals among new denture wearers. These results should suggest to all dentists that proper nutritional counseling is needed for the edentulous elderly in routine dental practice to prevent not only inadequate nutrition but also systemic diseases such as cerebrovascular disease.[8]

Muscle function is essential for masticatory efficiency. As individuals age, there is a significant decrease in the cross-sectional area and density of the masticatory muscles.[24] These observations are consistent with general age-related changes in skeletal muscle tissue throughout the body. Anthropometric measurements are indicators of muscle mass and subcutaneous fat.[25] Therefore, the significant decrease in AC and TSF observed among members of the lost contact group may also be associated with a reduction in masticatory function, resulting in increased difficulty chewing hard food. We did not evaluate such masticatory function elements as occlusal force and chewing efficiency in this study, however. Kikutani *et al.*[26] reported that oral function training can improve nutritional status in institutionalized elderly people. Further study will be needed to compare not only tooth contact but also masticatory function to evaluate the effects of nutritional status.

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臨床ヒント

新しい簡易口腔内細菌数測定装置の介護現場における臨床応用

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濱田　了[2,3)]・米山武義[4)]・菊谷　武[1,5)]

要旨：口腔内の環境および口腔ケアの質の評価をするために，口腔内に存在する細菌数を迅速かつ簡易に測定する細菌数測定装置を開発した．この機器を口腔ケアの現場で使用し，そのモニタリング機器としての有用性の確認と臨床応用の可能性を探ることを目的とした．

対象は，介護老人福祉施設の入居者41名（平均年齢84.5±8.2歳）である．対象に対し起床後・朝食前に舌背より細菌採取を行い，本装置を用いて口腔内細菌数の測定を行った．測定結果に基づき口腔ケアプランを策定し，口腔ケアを実施した．その結果，本装置で測定した舌上の細菌数の有意な変化が認められた（$p<0.05$）．以上より，本装置を口腔内汚染度のモニタリング機器として用いることは，口腔ケアを実施するにあたり，有用であることが示された．

Key words：Oral care, Oral bacteria, DEPIM（DiElectroPhoretic Impedance Measurement）

緒　　言

誤嚥性肺炎は，病原性微生物が誤嚥されることで引き起こされる気道感染[1)]であり，嚥下障害や胃食道逆流が起こるとそのリスクは高まるとされている[2)]．口腔内には，歯周病関連菌[3,4)]などの肺炎の原因となる細菌が多く存在する[5)]が，肺炎の原因菌を選択的に減少させることは困難である．Inglis ら[6)]は，肺炎発症には，誤嚥された細菌の種類より細菌の量に関係が深いことを報告している．専門的口腔ケアによって口腔内細菌数を減少させることが可能で[7,8)]，これにより，発熱日数の減少，肺炎の発症の抑制[9,10)]，さらには肺炎による死亡率が減少することが報告されている[11)]．

本研究で用いた機器は，誘電泳動とインピーダンス計測によるDEPIM（DiElectroPhoretic Impedance Measurement）法を応用したものであり，これまで著者らは本機器の精度の検討と臨床応用の可能性について検討してきた[12,13)]．

本研究では，この機器を用いて要介護高齢者に対する口腔ケアを行った際の効果を測定し，誤嚥性肺炎予防のための口腔内の汚染度のモニタリング機器としての有用性を検討した．

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対象および方法

1. 本研究に用いた機器

本研究に用いた機器はDEPIM法の測定に必要な要素から成り，細菌を捕集する電極チップおよび試料液を保持するセル，誘電泳動を誘起する交流電源回路およびインピーダンス計測回路などで構成されている．本装置はDEPIM法を利用することで簡易操作を実現しており，約5 m*l*の試料液および電極チップを装置にセットしてボタンを押す操作のみで測定が開始され，LCDに測定結果が表示される．

2. 口腔ケア介入と舌苔スコアおよび細菌数の変化

利用者の口腔ケアを口腔ケア・マネジメント[14,15)]に基づいて実施している介護老人福祉施設において，口腔ケアの効果を確認する目的で，本機器を用いて細菌数の測定を行った．対象は，某介護老人福祉施設に入居する要介護高齢者41名（男性7名，女性34名），平均年齢84.5±8.2歳とした．

口腔内細菌数の測定は，道重ら[16)]の報告に基づき，口腔内細菌数が最も多いとされる起床後の，水分を含めた経口摂取が行われる前とした．また，丸茂ら[17)]の報告に基づき，細菌数を最も多く認めた舌背後方部よりの採取とした．われわれの過去の研究において，舌より細菌を採取する際の圧力によって測定される細菌数に違いが生

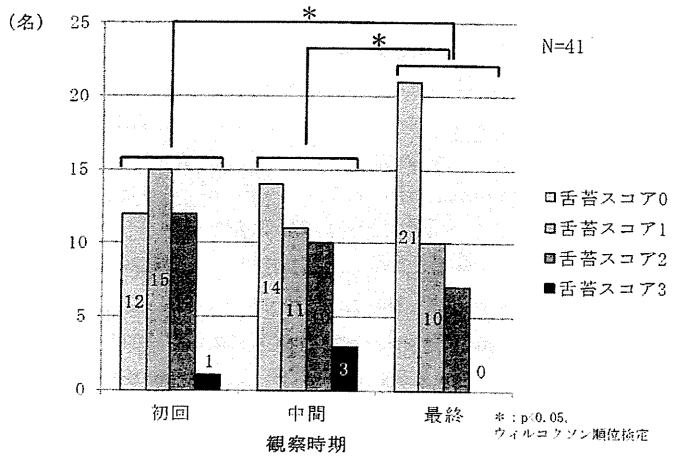

図1　観察期間中の舌苔スコアの変化

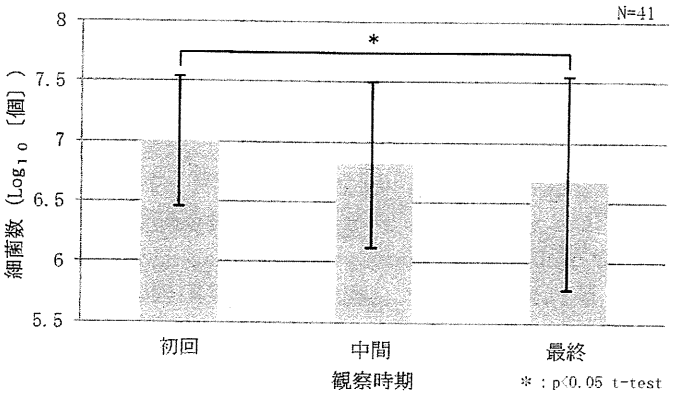

図2　観察期間中の細菌数の変化

じることを報告しており[18]，採取圧の誤差をなくすために採取圧を規定する装置を用い，舌背上約1cmの距離を滅菌綿棒を3往復させ擦過し検体とした．本機器による測定方法は上記に準じた．

さらに，舌苔付着状況をMiyazakiら[19]の基準に従い4段階で評価した．すなわち，舌苔スコア0：なし，舌苔スコア1：舌背の1/3未満が覆われているもの，舌苔スコア2：舌背の2/3未満が覆われているもの，舌苔スコア3：舌背の2/3以上が覆われているものである．細菌数の測定および舌苔スコアの評価は，6カ月にわたり行われた口腔ケアの実施期間のうち，初回，中間（3カ月後），最終（6カ月後）の3時点において行った．

口腔ケアは，週に1回2名の歯科衛生士が施設に出向き，口腔内環境の評価や自立度などの評価を行い，その結果に基づき口腔ケアプランを立案し，歯科衛生士みずからが口腔ケアを行うとともに，介護者に口腔ケアの指導を行った．残存歯は歯ブラシを用い，舌および粘膜はスポンジブラシを用いて剝離上皮や舌苔などの除去を行った．口腔ケア指導は，口腔内の衛生保持の必要性と本人の上肢機能などに合わせたセルフケアの方法について指導した．なお，中間時点において細菌数の変化から口腔衛生状態の改善が認められなかった者に対しては，セルフケア，介護者による口腔ケア，歯科衛生士による口腔ケアの割合の再検討や介護者への再度の口腔ケアの指導を行い，口腔ケアプランの見直しを行った．

統計学的検討は，SPSS Windows版Ver.16を用いた．舌苔の変化については，Friedman検定でデータ全体の有意の検定を行い，有意の場合には，Post-hoc testとしてBonferroni補正により得られたp値を基に有意水準5%としてWilcoxon順位検定を行った．さらに，細菌数の変化についてはt-testを用い，舌苔スコアと細菌数の比較はKruskal-Wallis検定を行い，有意水準5%未満を有意差ありとした．

本研究は，日本歯科大学生命歯学部倫理委員会の承認（07-06）を得て行われた．

結　　果

1. 口腔ケア介入と舌苔スコアおよび細菌数の変化

舌苔スコアは，初回2.02±0.84から3カ月後2.25±0.97，さらに6カ月後には1.61±0.77に変化した．Friedman検定によって有意な変化が認められ（$p<0.05$），3カ月後から6カ月後の間（$p=0.016$），初回と6カ月後の間（$p=0.031$）に有意差を認めた（図1）．

細菌数は初回7.00±0.54（Log_{10}〔個〕，以下同様）から3カ月後6.82±0.68，さらに6カ月後には6.67±0.90に変化した．初回から3カ月後には有意な変化が認められなかったものの，初回と6カ月後の間に有意な変化が認められた（$p=0.037$）（図2）．

舌苔スコアと測定された細菌数は，舌苔スコア0：平均細菌数6.68±0.81，舌苔スコア1：6.72±0.82，舌苔スコア2：6.80±0.66，舌苔スコア3：7.05±0.57となり，有意な関連を示さなかった（$p=0.299$）．

考　　察

要介護高齢者の重大な合併症である誤嚥性肺炎[20,21]は，口腔ケアによって予防できることが知られている[9,10]．要介護者や療養中の患者においては，みずからが，口腔内の環境を維持するための口腔の清掃を十分に行えない場合が多い[15]．そこで，これらの対象者の口腔内環境を維持するためには，歯科医師や歯科衛生士が口腔内環境を把握し，口腔ケアのプランを策定し，歯科スタッフ，介護スタッフや看護スタッフによる口腔ケアを実施しなければならない．有効なケアプランを策定し，実施するためには，対象者の口腔内状況を把握する必要

があると考える．

口腔ケアの指標として，プラークの付着度，舌苔の付着状況が指標として用いられ，臨床応用されている．Abeら[22,23]は，視覚的指標が発熱や肺炎の発症と関連があったことを示している．しかし，いずれも主観的な評価が中心で，客観的指標による評価はこれまであまり行われてこなかった．

舌背や義歯には多くの呼吸器感染症の原因微生物が存在することが知られている[5]．誤嚥性肺炎の原因菌は多岐にわたるため，誤嚥性肺炎の予防のための指標には，細菌の種類よりも細菌の数が重要であるとの報告もあり[6]，誤嚥性肺炎のリスクを評価するうえで細菌数の測定を行うことは意義のあることと考える．そこで著者らは，誘電泳動とインピーダンス計測によるDEPIM (DiElectroPhoretic Impedance Measurement) 法を応用し，検体採取から測定までに必要な時間は1分ほどで迅速に測定できる機器を開発した[12]．本機器の特性として生菌のみでなく死菌や食物残渣などの菌以外の物質も測定されるが，要介護高齢者を対象にした唾液をサンプルとした検討の結果，本機器による測定と従来法である培養法や蛍光法との相関は高いことを報告し，生菌数との高い相関が認められたことから，細菌数モニタリングとしての臨床応用が可能であると考えている[13]．そこで，本研究において，本機器を用いて，モニタリング機器としての有用性を検討することとした．その結果，口腔ケアの継続的な関わりのなかで，舌苔スコアの減少とともに本機器で測定した舌上の細菌数も減少を示した．また，3カ月後の中間地点で口腔内細菌数の減少をみなかった者も認められ，口腔ケアプランの見直しを行い実践した．

舌苔スコアと測定された細菌数は，有意な関連を示さなかった．舌苔スコアは舌苔の付着面積を基準とした指標であるのに対し，今回，細菌を採取した場所が舌の1カ所のみであったことが，両者の間に有意な関連が認められなかった原因と考えられる．しかし，口腔ケアの介入によって，舌苔スコアおよび細菌数ともに有意な低下を示したことから，本装置で測定された細菌数を口腔ケアの指標として用いることは妥当性があると考える．

口腔ケアの質の管理には，その実施にあたり，「対象者の口腔内汚染度を評価し，その結果を口腔ケアプランに反映させて，口腔ケアを実施し，その後に口腔ケアの効果を確認し，さらにその結果をケアプランに反映させる」といった，PDCAサイクルに則った口腔ケアのシステムが推奨される[14,15]．本機器のもつ簡易で迅速に測定できるという特徴は，口腔ケアの質を維持するためのシステムを実施するにあたり，有用であると考えられた．

結　論

要介護高齢者に対し，歯科衛生士による口腔ケアの介入により舌背スコア，細菌数に変化が認められた．口腔ケア介入効果の判定に細菌数を用い，その測定に結果が迅速に判明する本機器を用いることは有用であることが示唆された．

利益相反の開示

本論文の研究に使用した簡易口腔内細菌数測定装置は，株式会社パナソニックヘルスケアから貸与を受けたものである．

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Development and Clinical Application of a Novel Rapid Oral Bacteria Detection Apparatus

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A rapid oral bacteria detection device was newly developed for the evaluation of oral hygiene and the quality of oral health care by measuring the amount of oral bacteria. The aim of this study was to explore the potential clinical utility for oral health care as checking and monitoring equipment.

The subjects were 41 elderly residents (7 men and 34 women) aged 84.5±8.2 years old (mean age±SD) in a welfare facility for the elderly and requiring long-term care. The amount of oral bacteria was measured using the device from the dorsum of the tongue, in the morning before breakfast. Based on the results, the dental hygienists prepared an interventional oral health care plan for the subjects, and conducted oral health care intervention during 6 months.

The results of this study showed that there was significant changes in the amounts of oral bacteria on the tongue surface measured by this device ($p<0.05$). Use of this device was shown to be potentially useful as a monitoring device when performing oral health care.

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口臭症の基礎知識：定期受診は歯科を活性化する

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歯科界危機の主因は，極めて低い定期受診率にある。著者の研究では人々が口臭症の害を認識することで，定期受診が普及し公衆衛生改善が期待できる。その社会医学論理と基礎知識を概説する。

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はじめに

わが国歯科界は未曽有の危機に立たされている。歯科大学も同様で，昭和40年代のように優秀な受験生が先を競った時代とは大きく様変わりをした。黄金期を知る開業医・大学人にとって今は，「理想」と葛藤する毎日であろう。こういう時代こそ，高度経済成長期直前（昭和30年代初期）の大不況が教えるように，発想を逆転する必要がある[1]。すなわち，保険点数や国試合格率等にとらわれることなく，10年先を想像し，５年先を予想する「一貫したポリシー」が必要となる。

歯科界衰退の原因と対策

著者は北米で臨床の大学教授と開業医を経験した。そのためか，歯科界衰退の原因が一目瞭然に見えてしまう。北米人に著者の仮説「歯科定期受診の圧倒的不足が日本歯科界衰退の原因」の正否を問うと，皆「今さら何を言っているのだ？　当り前だろう」と返してくる。行うべきは，歯科定期受診・予防（クリーンアップ）の普及である。第２は低い診療費の改善，第３は治療内容の一層の近代化となる。しかし，この３つを一挙に解決するのは，日本の経済状況，社会が許容しない。先ずは定期受診から手を付けるべきであろう。**歯を削らず国民の健康を守る「大きな業績」**に国民が納得してくれれば，低歯科診療費の問題も解決できるであろう。

ところが今，日本で流行し始めた「予防」は少々違う。世界の予防は，社会医学に立脚した真の予防である。その結果，「国民の歯科界への価値観」が向上し，歯科医は世界で最高の職業となった。またイギリスの先例では実に１/３の国民が，同国が誇る健康保険治療ではなく「より充実した高サービスの自由診療」を求める社会となった[2]。

一方，口臭症の罹患率は50％を超えると推定され[3]，歯科症状の中では「冷水痛」に次ぎ多い[4]。齲蝕・歯周病を超えるが，歯科を受診する患者は少ない。著者の研究によれば，口臭症予防の推進は，オーラルヘルスプロモーションに繋がり歯科受診率の増加が期待できる。そのうえ，口臭は歯周病原性のみならず発癌性や幹細胞への毒性，全身毒性がある[5~11]。これらの毒性は，国民には意外性のある事実だ。すなわち，定期受診を勧める患者教育教材としては，口臭症は最適に近い。

ところが，なぜかEBMのない口臭治療法・診断法が日常臨床に氾濫しているため，歯科界は口臭臨床を大きく誤解している[12]。そして，口臭に目を向ける者が少なく，本稿で示す「口臭臨床の重大な役割」には気づいていない。すなわち口臭の正しい知識を知る必要がある。

ARTICLES

大きな誤解：予防と歯科医業収入

歯科界では，「予防の目的」が十分理解されず[13]，定期受診・予防は患者を減らすかのように誤解されている。しかしアメリカの先例では定期受診・予防が普及した30年ほどで，国民歯科医療費の対GNP比は驚くべき事に２倍近くも増加した[14]。日本と同じく，健康保険のあるイギリスでも，定期受診する者では残存歯が約２本/５年と増えて公衆衛生に貢献する一方，修復歯面数は僅か５年で２倍も増加した[15]。この結果は欧米の他の多くの報告が支持している[16]。

日本の予防歯科臨床は地域歯科保健との連携が少ない。そしてパターナリズムの濃い予防歯科臨床になっている。また多くが，メインテナンスのレベルに留まり，予防の意味が充分理解されていない[13]。口下手な臨床医は「今流行の予防歯科」に失敗する。その一方，カリスマ性のある歯科医は大成功する。このような現象は公衆衛生学的失敗である。大学で教える予防歯科では，「地域との連携」と「１次予防・２次予防の概念」が必須であり，カリスマ性は必要でない[13]。欧米のように歯科界が患者を「コンシューマー」と見なし公衆衛生的立場から動向を見る必要がある。そして，国民を定期受診に誘導する姿勢が求められている[17]。実は，この目的に口臭臨床が使えるのである。

歯科医過剰と定期受診

表１に，先進各国の人口当たり歯科医師数を示した。歯科界の思いとは異なり，日本の歯科医師数は少なめである。また日本の歯科医師は「齲蝕が減ったから患者が減った」と言う。**図１**に示したが，ここに大きな誤りが２点ある。１．一人あたりの永久歯齲蝕は2005年まで着実に増加している（**図１**）[18~21]。２．減っているのは乳歯と12歳児の永久歯齲蝕。では，なぜ「歯科医師過剰」を声高に叫ぶ必要があるのか。これを考えたい。

アメリカの歯科医師数は，**表１**に示した数よりずっと多いと見積もられる。一方，日本では，あまり臨床を行わない高齢歯科医あるいは準専業主婦歯科医も統計に数えられている。したがって実動数はアメリカより少ない。ところがアメリカでは15年前より歯科医不足が始まった[22]。そこで毎年，千人規模で外国人歯科医師に歯科医師免許を与えている。しかし，それでも歯科医不足は深刻で，過去２－３年で歯学部が２つ増設された。アメリカの大都市しか知らない日本人歯科医には意外な話だ。

それに対し，齲蝕が増えている日本では歯科医過剰を叫んでいる。もちろんDMFTなので「実態に合わない」との声もある。しかし，この意見は公衆衛生学が歯科界に根付いていない証拠でもある。「歯を削る」だけが歯科の仕事では無い[23]。

歯科医の責務を「公衆衛生への貢献（歯科医師法第一条）」と知る歯科医には，「北米の現状」に大きな期待を抱くであろう。すなわち「定期受診・予防歯科の一般化」で国民の公衆衛生の向上・増進に大いに貢献

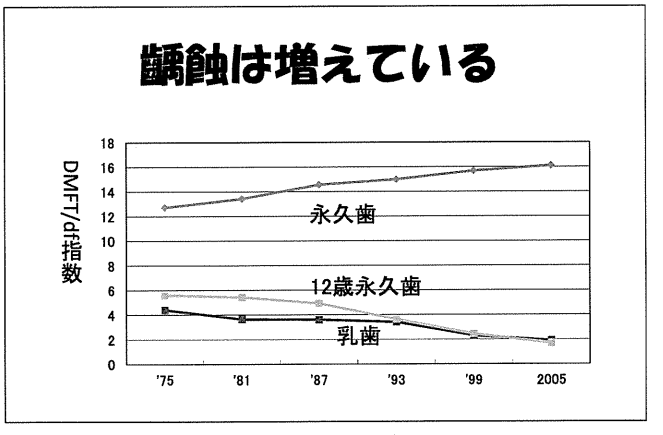

図１　日本人のむし歯の数（1957年以降）[18~21]
一般に齲蝕は減って来たと言われるが，それは乳歯と12歳児のDMFTであり，日本国民全体でみると未だ増加傾向を示している。

表１　各国の歯科医師数（人口１万人当たり）[13]

国名	日本	アメリカ	デンマーク	ドイツ	イタリア	フランス	ベルギー
歯科医師数	7.6	6.1	9.2	8.1	8.1	6.4	7.1
調査年	2008	2007	2009	2008	2007	2008	2007

アメリカは歯科医師会調査のため実数を表していない。一説には「10人に近い」との話も聞かれる。

するだけでなく，一般歯科臨床が活性化するからである。「定期受診・予防歯科」で歯科界は再生する。**表2**に示すように，定期受診率が北米なみとなれば，一挙に歯科医は不足する[23]。

日本歯科大学の創設者・中原市五郎先生は昭和初期，著書である「日本食養道」の中で「歯科医院は予防医学の中心たれ」と言葉きつく強調している。食育中心の思想ながらも80年前から中原市五郎先生は，予防医学の重要性を歯科医師に教えている。歯科界が，中原先生の言に従っておれば，今の状況は無かったであろう。

予防リコールとメインテナンスは違う。メインテナンスは3次予防的要素が多く1次予防的要素は少ない。したがって対象者が少ない[13]。予防リコールを成功させるには，「地域歯科保健との連携」が不可欠である[13,25]。これは，臨床医自らが作る連携であり，国・地方公共団体の仕事では無い。国民から歯科保健に理解を得て，歯科定期受診を推進するには，歯科界の努力が必要である。

「地域歯科保健との連携」を行うには，人々の認識を得るための社会活動（健康教育では無い）が先ず必要となる。これなしに一足飛びに，健康教育を行うのは社会医学的誤りである。表現が不適切だが，この目的には広告塔が必須である。広告塔として「口臭臨床」は大いに役立つ。

定期受診の推進：口臭臨床の大いなる価値

成人の約半分に口臭があるはずだが[3]，女子大生を調査すると，自分の口臭を気にする者は2割未満と少ない。しかし，「他人の口臭を気にする者」は2倍に増える[26]。若い女性は自分に自信があるのだろう。ところが自分の口臭には気づいていない。もちろん，対人関係への悪影響にも気づいていない。これは口臭予防を勧める材料となる。

ジョンソン＆ジョンソン日本法人ヤンセンファーマ社が，OLを対象に「理想的な上司でも身だしなみが気になるか？」と訊くと9割が気になると言う。それも**図2**に示すように，最も気になるのは「口臭・体臭」である。一流企業のサラリーマン：日本の時流を創る人々に，口臭は，定期受診推進の大きな説得材料となる。もし彼らを予防歯科に取り込めたら，定期受診は，自然に日本の習慣となる。

ライオンが日米のサラリーマンで，定期受診率を比較してみた。すると日本で定期健診しない者が圧倒的に多い（**図3**）。また歯に自信が無い者も非常に多く，理由は口臭だという（**図4**）。すなわち口臭予防は，サラリーマンの定期受診の動機となる。著者の研究でも，通常の保健教育に比べ，口臭教育を行うと「定期受診する」という者が増加した。

子供の定期受診推進については，北米では確立した手法がある[27]。わが国では，子供だけでなく，成人にも定期受診を促進しなければならない。歯科医には，国民を予防歯科に取り込む十分な知識とレトリックが

表2　歯科医師数と歯科医院定期健診の習慣[4,24]

	歯科医師（人口1000人）	定期受診率
日本	0.76人	14 %
北米	0.61人	68 %

（OECD・カナダ歯科医師会調査を含む）

日本の定期受診は期間を限らず，北米では1年に1回以上である。北米の歯科医師の実数は，本表に挙げた数より随分多い。一方，日本の実動数は表に示すより少なく，北米より少ない可能性が高い。北米なみに定期受診が増えれば，一挙に歯科医師不足となる。

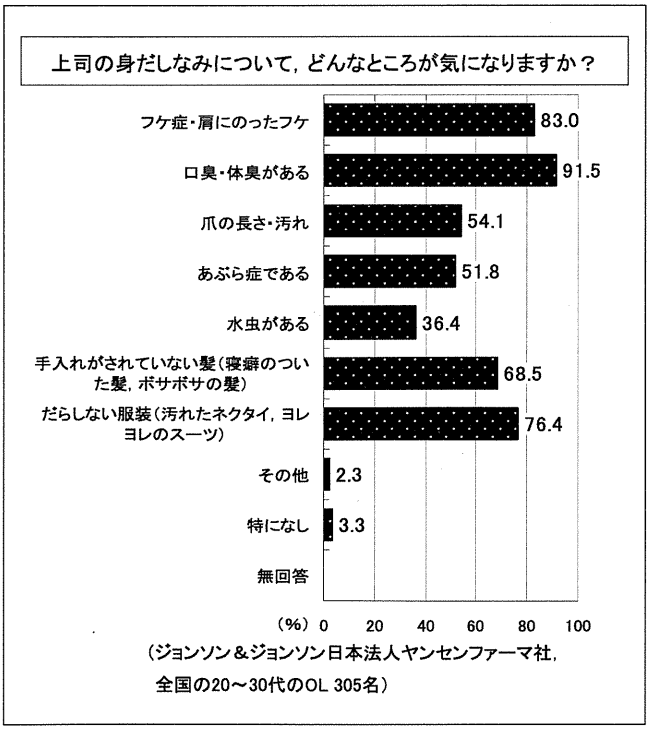

図2　ヤンセンファーマがOLを対象に「理想的な上司でも，気になる身だしなみは何か」と訊いてみた。トップは「口臭・体臭がある」であった。部下を持つビジネスマンは，口臭を予防しなければ，部下OLとの関係に支障が出てくることを知ると受診の動機づけになる。