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Sterilization effects of commercial denture cleaners compared with a combination of denture cleaners and ultrasonic cleaning

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We evaluated the sterilization efficacy of a commercial denture cleaner compared with the combined use of the denture cleaner and ultrasonic cleaning on dentures used by patients. After cleaning the dentures with a denture brush, bacteria were collected from one half of the mucosal surface with a sterile cotton swab. Complete and partial dentures were cleaned by immersion in a dedicated commercial denture cleaning solution for 30 min. We also examined the effect of 5 min of cleaning with a commercial ultrasonic cleaner and a denture cleaner after mechanical cleaning. The dentures with a viable bacteria count greater than 4.54×10^9 CFU/mL before rinsing at 40°C showed a high sterilization rate greater than 95%. The sterilization rate of dentures with a low bacterial count before immersion was low. Similar results were obtained for anaerobic bacteria. Regarding the ultrasonic cleaner, debris was visually removed from all 30 dentures. These results suggest that it is important to evaluate denture cleansers to the extent that they can reduce bacterial counts rather than sterilization rates. (J Osaka Dent Univ 2020 ; 54 : 225-238)

Key words : Denture cleaner ; Sterilization ; Ultrasonic cleaner

INTRODUCTION

According to the “Survey of Dental Diseases,”[1] in Japan, the number of teeth in the elderly has increased because of improved public awareness of oral health, with 51.2% of those 80 years or older having more than 20 teeth (the so-called “80/20” goal). However, due to the increase in the aged population, there has been little decrease in the use of dentures. According to the “Survey of Dental Diseases,”[1] more than half of those over 55 years have dental prostheses. For those under 85 years of age, most people have bridges on the missing teeth. On the other hand, for those over 85 years of age, there are as many with fixed prostheses as with removable prostheses. The number with removable prostheses increases with age. Therefore, the maintenance of dentures remains an important issue.

Denture plaque with a thickness of 4-30 μm is deposited on the surface of the dentures. It is not a mere microbial clump, but rather, a biofilm in which microorganisms, saliva, and serum are aggregated. Denture plaque is defined as “a biofilm containing 10^{11} to 10^{12} microorganisms/g wet weight formed on the surface of dentures.”[2] It is well known that microorganisms in denture plaque are associated with systemic infections such as aspiration pneumonia, enteric infections, meningitis, and infective endocarditis, as well as dental caries, periodontal disease, and oral mucosal disease.[2, 3] Therefore, the sterilization of dentures by removing denture plaque is very important for maintaining not only oral, but also general health. Denture cleaning is mainly performed by mechanical removal with a denture brush and chemical sterilization with a denture cleaner. It is said that although cleaning with a denture brush is effective for denture plaque that can

be seen with the naked eye, it is less effective for plaque that cannot be seen.[4] Therefore, chemical cleaning with denture cleaners is important for denture sterilization.

Many studies have evaluated the sterilization effect of denture cleaners *in vitro*[5] or with microorganisms that experimentally adhere to resin surfaces.[6] Although the results of these studies have verified the efficacy of denture cleaners, few studies have reported their bactericidal effect on dentures that are actually in use. In this regard, Inoue *et al.*[7] found that denture cleaners are effective for denture sterilization, and that when using denture cleaners, warm water is more effective than room temperature water for sterilization. Recently, ultrasonic denture cleaners that can be used at home have become commercially available. For many elderly who use dentures, especially those in care facilities, it is difficult to clean mechanically with a denture brush. An ultrasonic cleaner may be most effective. The use of denture cleaning solution and ultrasonic cleaning is considered capable of achieving a very high disinfection effect.[8, 9] However, the sterilization effect on dentures that are actually being used has not been sufficiently verified. Examining dentures actually used by patients, we studied the sterilization efficacy of a commercially available denture cleaner, as well as a combination of the denture cleaning solution and ultrasonic cleaning.

MATERIALS AND METHODS

We examined complete dentures and bilateral extension removable partial dentures actually used by 53 patients (19 males and 34 females) with an average age of 74 years who were seen by the Department of Fixed Prosthetics and Occlusion or the Department of Geriatric Dentistry at Osaka Dental University Hospital, or the Yagi Dental Clinic. We excluded those who were using antibacterial agents in their oral cavity, soft lining materials, or denture stabilizers, as well as those who were objectively judged to lack the ability to provide informed consent. Figure 1 shows the denture cleaning brush (Kobayashi Pharmaceutical, Osaka, Japan). Figure 2 shows the mechanical cleaning method before immersion in the denture cleaner. The denture was brushed about three times to remove saliva and debris on the mucosal surface. Figure 3 shows the collection of the bacteria adhering to the denture before it is washed with the denture cleaner. One side of the mucosal surface of the denture was rubbed twice with a sterile cotton swab to collect the bacteria before cleaning. The bacteria were stored in an iSWAB® storage device (Mawi DNA Technologies, Houston, TX, USA). Figure 4 shows the cleaning method with a denture cleaner. Complete and partial dentures were immersed for 30 min in a cleaning solution made by dissolving Tough Dent™ or Partial Dent™ (Kobayashi Pharmaceutical, Osaka, Japan) in 200 mL of water in a special cup at 40°C or 15°C. Ultrasonic Tough Dent™ was used for the combination method with an ultrasonic cleaner. The dentures were immersed in a cleaning solution of a special cleaning agent

Fig. 1 The denture brush

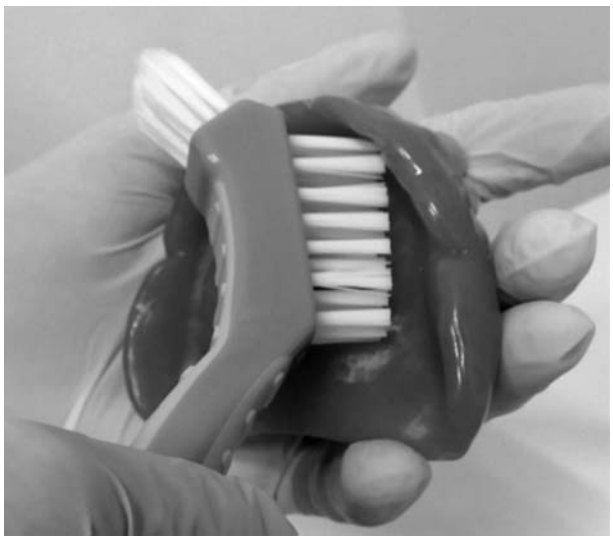

Fig. 2 Cleaning method with the denture brush. The mucosal surface was brushed with three strokes under running water.

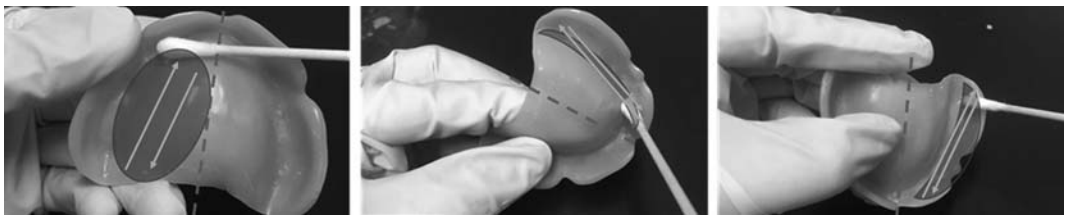

Fig. 3 Collection of bacteria adhering to the denture before washing with the denture cleaner. The three locations shown were rubbed twice with the same swab. The collected bacteria were stored in a storage device.

A B

Fig. 4 Denture cleaning methods. (A) With the immersion method the denture is placed in a storage cup with the cleaning solution at 40°C or 15°C for 30 min. A tablet for either a complete (left) or partial (right) denture is dissolved in water to make the solution. (B) With the commercial ultrasonic denture cleaner, the prosthesis is immersed in a solution at 15°C for 5 min.

dissolved in 140 mL of water at 40°C for 5 min to carry out the ultrasonic cleaning according to the manufacturer's instructions.

After removal the denture, the cleaning solution on the denture surface was washed off with tap water. Thereafter, in the same method as that before cleaning, a sterile cotton swab was drawn across the mucosal surface of the denture twice on the opposite side from which the bacteria were collected before cleaning, to collect the bacteria. The bacteria were then stored in the same storage device with those from before cleaning. The stored bacteria were spread on Soybean-Casein Digest (SCD) agar medium for aerobic bacteria (Fujifilm Wako Pure Chemical, Osaka, Japan), modified Gifu Anaerobic Medium (GAM) agar for anaerobic bacteria (Nissui Pharmaceutical, Tokyo, Japan), X-SA medium for staphylococci (Nissui Pharmaceutical, Tokyo, Japan), sheep blood agar medium for streptococci (Nissui Pharmaceutical) and Candida EX medium for *Candida* (Nissui Pharmaceutical). They were then subsequently cultured. The viable bacteria counts were measured by sending samples to Kobayashi Pharmaceutical, or Kitakyushu City Pharmacists Association Inspection Center, Kitakyushu, Japan.

Based on the viable counts of the aerobic bacteria, anaerobic bacteria, staphylococci, streptococci, and *Candida* before and after cleaning, the sterilized bacteria count and sterilization rate were determined for each cleaning method. Overall differences between the three methods were determined by one-way analysis of variance (ANOVA). If the one-way ANOVA was significant, differences between cleaning methods were estimated using Fisher's least significant difference (LSD) test. This study was approved by the Osaka Dental University Medical Ethics Committee (Approval No.111004).

RESULTS

Differences in sterilized bacteria counts for each cleaning method

Table 1 shows the differences in sterilized viable bacteria counts for each cleaning method. ANOVA indicated that cleaning significantly reduced the sterilized bacteria count of aerobic bacteria ($p < 0.01$), as well as both anaerobic bacteria and streptococci ($p < 0.05$). Regarding the aerobic bacteria, Fisher's LSD test indicated that cleaning at 40°C reduced the sterilized bacteria count significantly more than cleaning at 15°C or cleaning with the combined use of the ultrasonic device ($p < 0.01$). Regarding the anaerobic bacteria, cleaning at 40°C reduced the sterilized bacteria count signifi-

Table 1 Differences in sterilized bacteria count by cleaning method.

	Cleaning conditions	Number of samples	Sterilized bacteria count(CFU/mL)	
Aerobic bacteria	Cleaning solution at 40°C	22	$7.5\times10^{9}\pm1.2\times10^{10}$	*, **
	Cleaning solution at 15°C	14	$-6.6\times10^{6}\pm2.2\times10^{7}$	
	Cleaning solution at 40°C and ultrasonic device	30	$7.9\times10^{6}\pm3.1\times10^{7}$	
Anaerobic bacteria	Cleaning solution at 40°C	22	$3.4\times10^{9}\pm8.8\times10^{9}$	*
	Cleaning solution at 15°C	14	$-4.4\times10^{8}\pm2.8\times10^{7}$	
	Cleaning solution at 40°C and ultrasonic device	30	$5.8\times10^{6}\pm2.0\times10^{7}$	
Streptococci	Cleaning solution at 40°C	23	$4.1\times10^{9}\pm9.4\times10^{9}$	*, **
	Cleaning solution at 15°C	14	$-4.1\times10^{5}\pm1.3\times10^{7}$	
	Cleaning solution at 40°C and ultrasonic device	30	$6.7\times10^{6}\pm4.9\times10^{7}$	
Staphylococci	Cleaning solution at 40°C	20	$5.4\times10^{4}\pm2.5\times10^{6}$	
	Cleaning solution at 15°C	8	$-6.6\times10^{5}\pm1.4\times10^{6}$	
	Cleaning solution at 40°C and ultrasonic device	8	$1.1\times10^{6}\pm2.9\times10^{6}$	
Candida	Cleaning solution at 40°C	20	$7.0\times10^{5}\pm2.1\times10^{6}$	
	Cleaning solution at 15°C	9	$-6.3\times10^{4}\pm1.3\times10^{6}$	
	Cleaning solution at 40°C and ultrasonic device	10	$1.4\times10^{5}\pm2.5\times10^{5}$	

The sterilized bacteria count is the number of viable bacteria before cleaning minus the number after cleaning. Mean ± SD, $^{**}p<0.01$, $^{*}p<0.05$.

cantly more than cleaning with the combined use of the ultrasonic device ($p<0.05$). Regarding streptococci, cleaning at 40°C reduced the sterilized bacteria count significantly more than by either cleaning at 15°C ($p<0.05$) or by the combined use of the ultrasonic device ($p<0.01$). Regarding staphylococci and Candida, no significant differences were seen in the sterilized bacteria count among the cleaning methods.

Relationship between the viable bacteria count before and after cleaning and the sterilized bacteria count and rate

Cleaning with the denture cleaning solution at 40°C

Figure 5 shows that the relationship between the viable bacteria counts before and after cleaning with the denture cleaner solution at 40°C. The viable counts of aerobic, anaerobic, and streptococcal bacteria after cleaning were small, regardless of the value before cleaning. Because of the low viable count, the maximum values on graphs of staphylococci and *Candida* are 1/1000. No significant differences in viable bacteria counts were observed before or after cleaning for these bacteria, and there was little sterilization effect. Figure 6 shows the relationship between the sterilization rate and viable bacteria count before cleaning with the denture cleaner solution at 40°C. Although the eradication rate of the aerobic, anaerobic, and streptococci bacteria was uneven in dentures with a low viable count before cleaning, the sterilization rate increased to nearly 100% as the viable count increased. Because of the low viable count, the maximum value on graphs of staphylococci and *Candida* was 1/1000. No clear relationship was observed between the sterilization rate and viable count before cleaning for these bacteria. As shown in Figs. 5 and 6, the high sterilized bacteria counts for the aerobic, anaerobic, and streptococci bacteria after cleaning at 40°C (Table 1) was found to be related to the high viable bacteria count before cleaning. No significant difference in the sterilization effect was found between complete and partial dentures.

Cleaning with denture cleaning solution at 15°C

Figure 7 shows the relationship between the viable bacteria counts before and after cleaning with the denture cleaner solution at 15°C. Because the viable bacteria counts of the aerobic, anaerobic, and streptococci bacteria were lower than those after

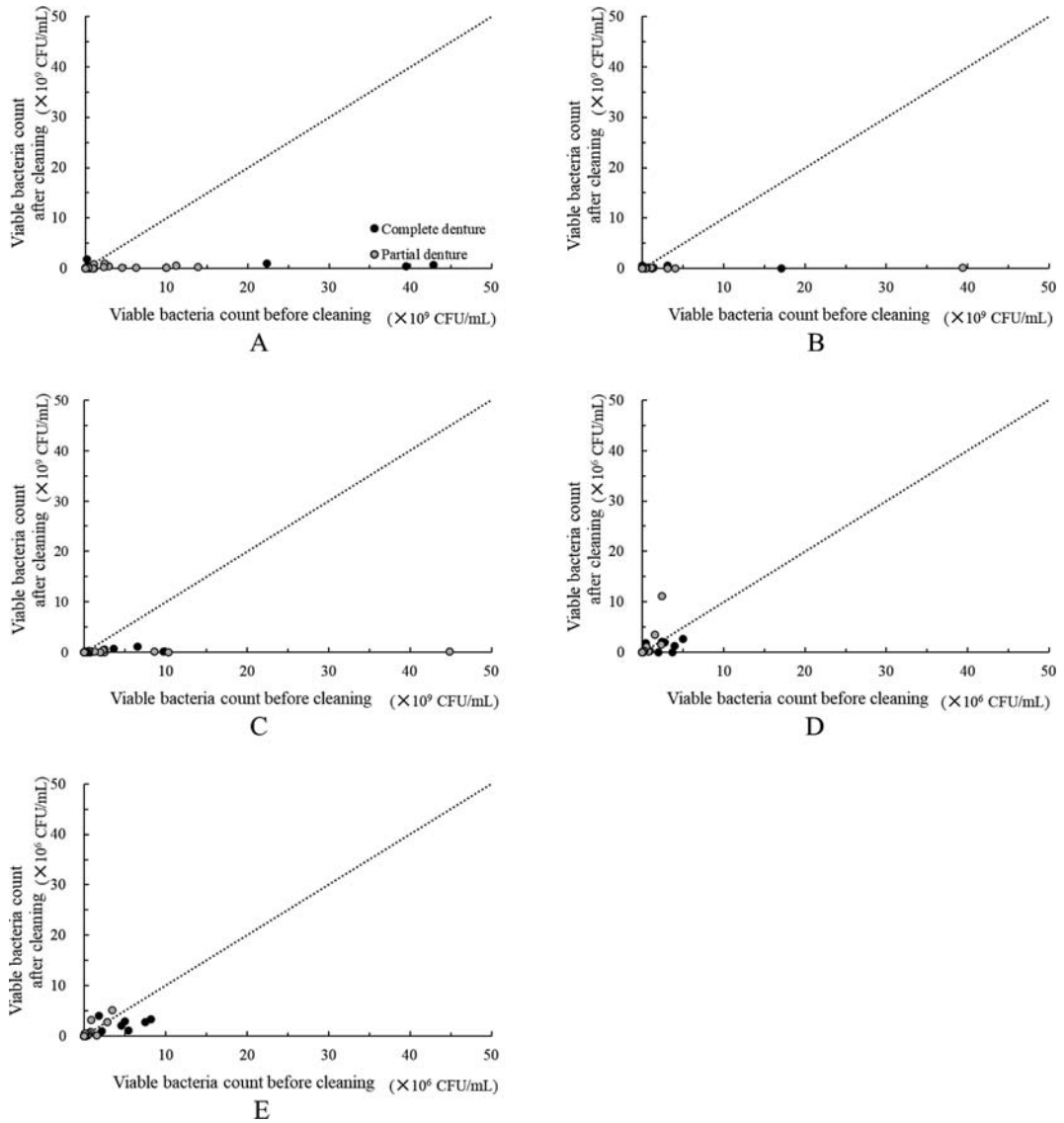

Fig. 5 Relationship between the viable bacteria counts before and after cleaning with the denture cleaner solution at 40°C for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.* The dotted lines represent change in the viable bacteria count before and after cleaning.

cleaning at 40°C, the maximum values on graphs were 1/100 of those shown in Fig. 5. No significant differences were observed in the viable bacteria counts before and after cleaning at 15°C, and hardly any sterilization effect was seen. The maximum values on graphs for the staphylococci and *Candida* were further reduced to 1/100 of those described above for the three bacterial species because the viable cell count was even lower. No significant difference in the bacteria count before and after cleaning was observed. Moreover, some dentures showed an increased viable count after cleaning.

Figure 8 shows that the relationship between the sterilization rate and viable bacteria counts before cleaning with denture cleaner solution at 15°C. Because of the low viable bacteria counts, the maximum values for the aerobic, anaerobic, and streptococci bacteria were 1/100 of those shown in Fig. 6. In these bacteria, no clear relationship was ob-

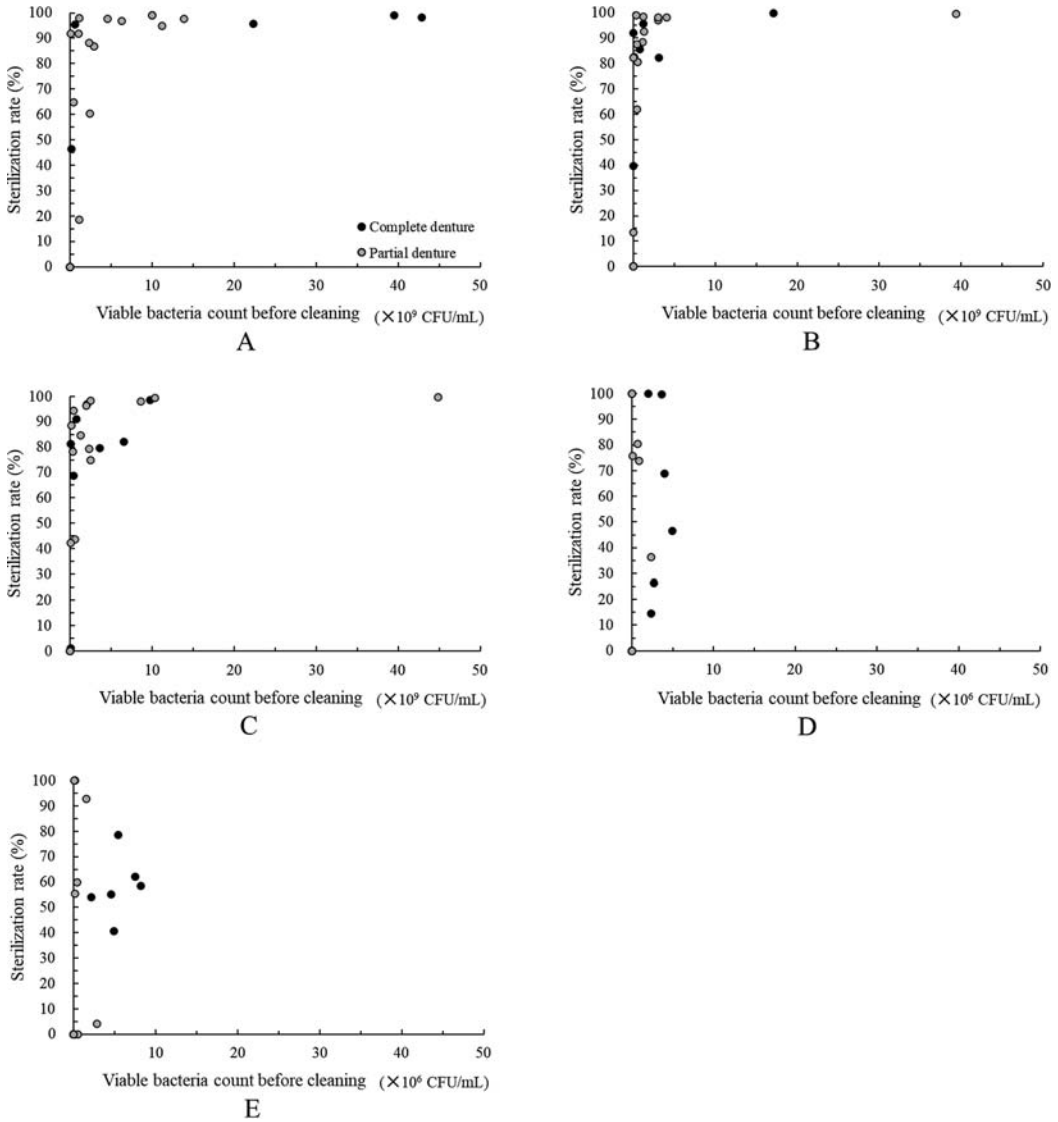

Fig. 6 Relationship between the sterilization rate and viable bacteria count before cleaning with the denture cleaner solution at 40°C for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.*

served between the sterilization rate and the viable count before cleaning. The maximum values on graphs of staphylococci and *Candida* were further reduced to 1/100 of those described above for the three bacterial species because the viable cell count was even lower. Staphylococci and *Candida* were only detected on a few dentures before cleaning. Some dentures on which *Candida* was detected showed a sterilization rate close to 100%.

Based on the results shown in Figs. 7 and 8, the low sterilized bacteria counts of the aerobic, anaerobic, and streptococci bacteria after cleaning at 15°C were related to the low viable bacteria count before cleaning, and hardly any sterilization effect was observed when the viable bacteria count was low before cleaning. Similar to the cleaning at 40°C, little if any difference was observed in the sterilization effect between the complete and partial dentures.

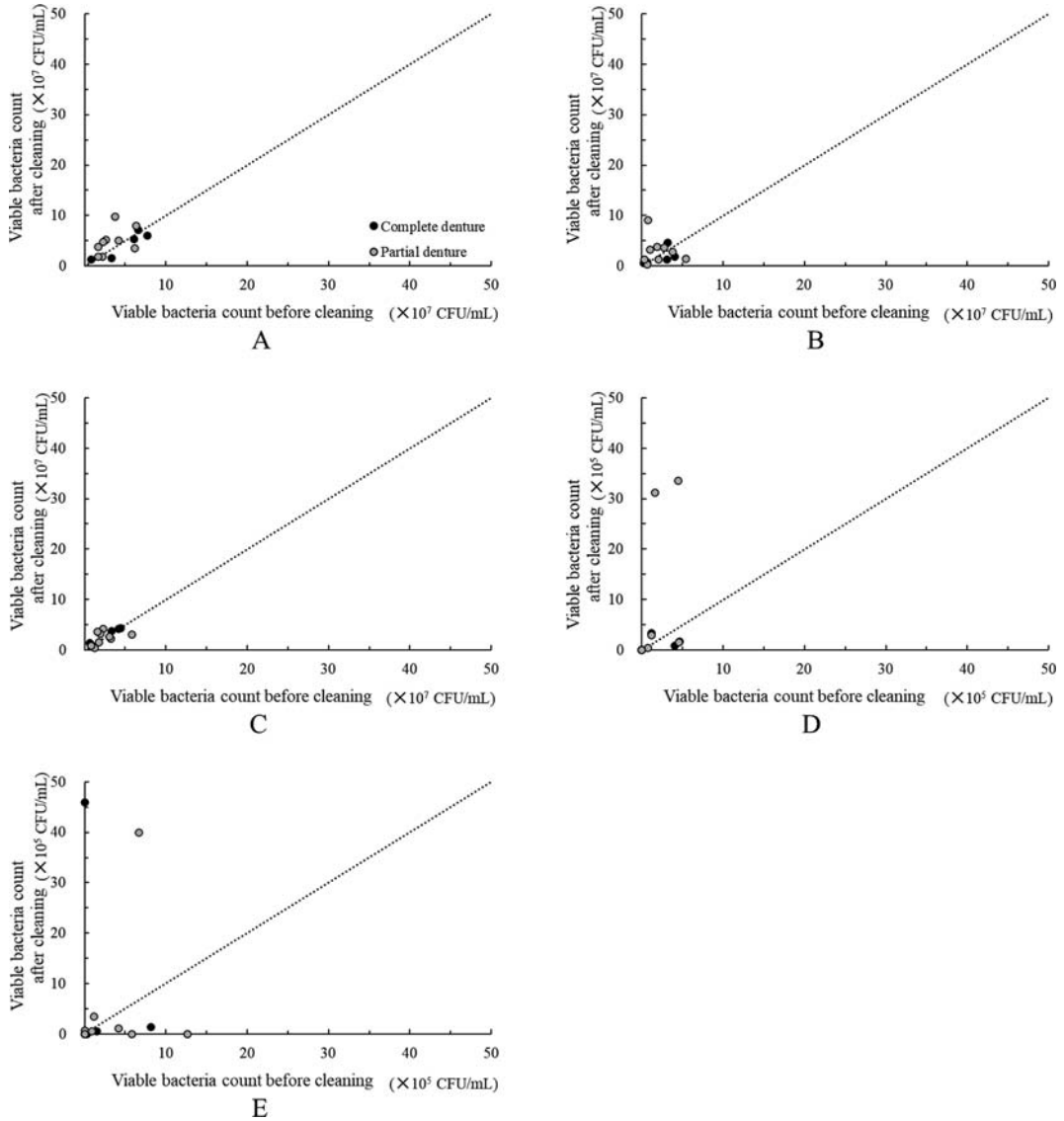

Fig. 7 Relationship between the viable bacteria count before and after cleaning with the denture cleaner solution at 15°C for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.* The dotted lines represent change in the viable bacteria count before and after cleaning.

Combined use of denture cleaning solution and the ultrasonic device

Figure 9 shows the relationship between the viable bacteria count before and after cleaning with the combined use of the denture cleaning solution and ultrasonic device. Similar to the cleaning at 15°C, because the viable bacteria counts of the aerobic, anaerobic, and streptococci bacteria were lower than those after cleaning at 40°C, the maximum values on the graphs shown in Fig. 9 are 1/100 of those shown in Fig. 5. No significant differences were seen in the viable bacteria counts before and after cleaning, and there was hardly any sterilization effect. Due to the low viable bacteria count, the maximum values on graphs of staphylococci and *Candida* were further reduced to 1/100 and 1/10 of the above three bacteria species, respectively. Even for these bacteria, no significant difference was found in the viable bacteria counts before and after cleaning, and there was hardly any steriliza-

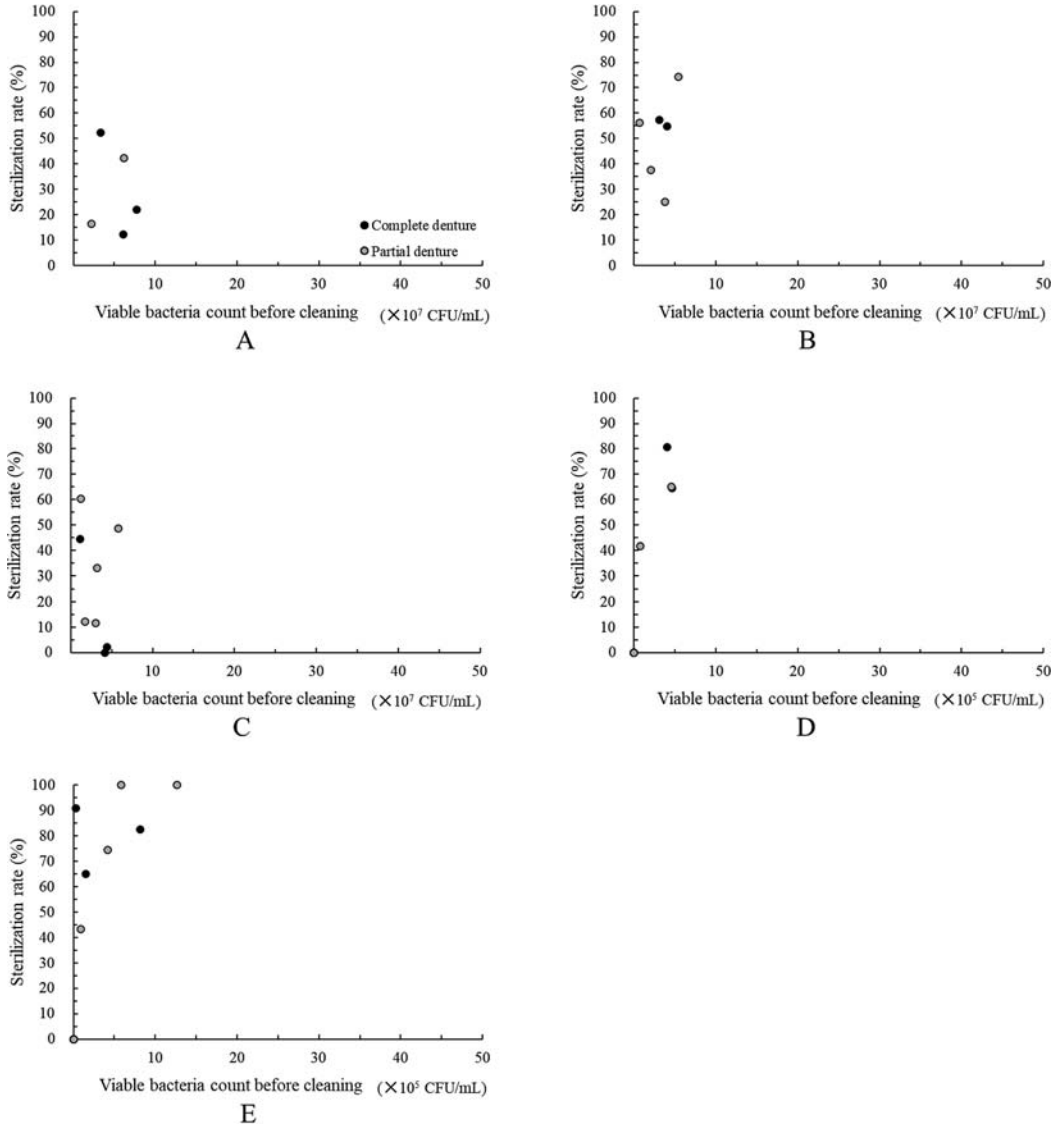

Fig. 8 Relationship between the sterilization rate and viable bacteria count before cleaning with the denture cleaner solution at 15°C for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.*

tion effect.

Figure 10 shows the relationship between the sterilization rate and viable bacteria counts before cleaning with the combined use of the denture cleaning solution and the ultrasonic device. Due to the low viable bacteria count, the maximum values on graphs of the aerobic, anaerobic, and streptococci bacteria were 1/100 of those shown in Fig. 6. For these bacteria, no significant differences were found in the viable counts before or after cleaning because many dentures had a low viable bacteria count before cleaning. However, a tendency was observed for the sterilization rate to increase with increases in the viable bacteria count before cleaning.

Due to the low viable bacteria count, the maximum values on graphs of staphylococci and *Candida* were further reduced to 1/100 and 1/10 of the above three bacteria species, respectively. Staphylococci and *Candida* were detected on fewer den-

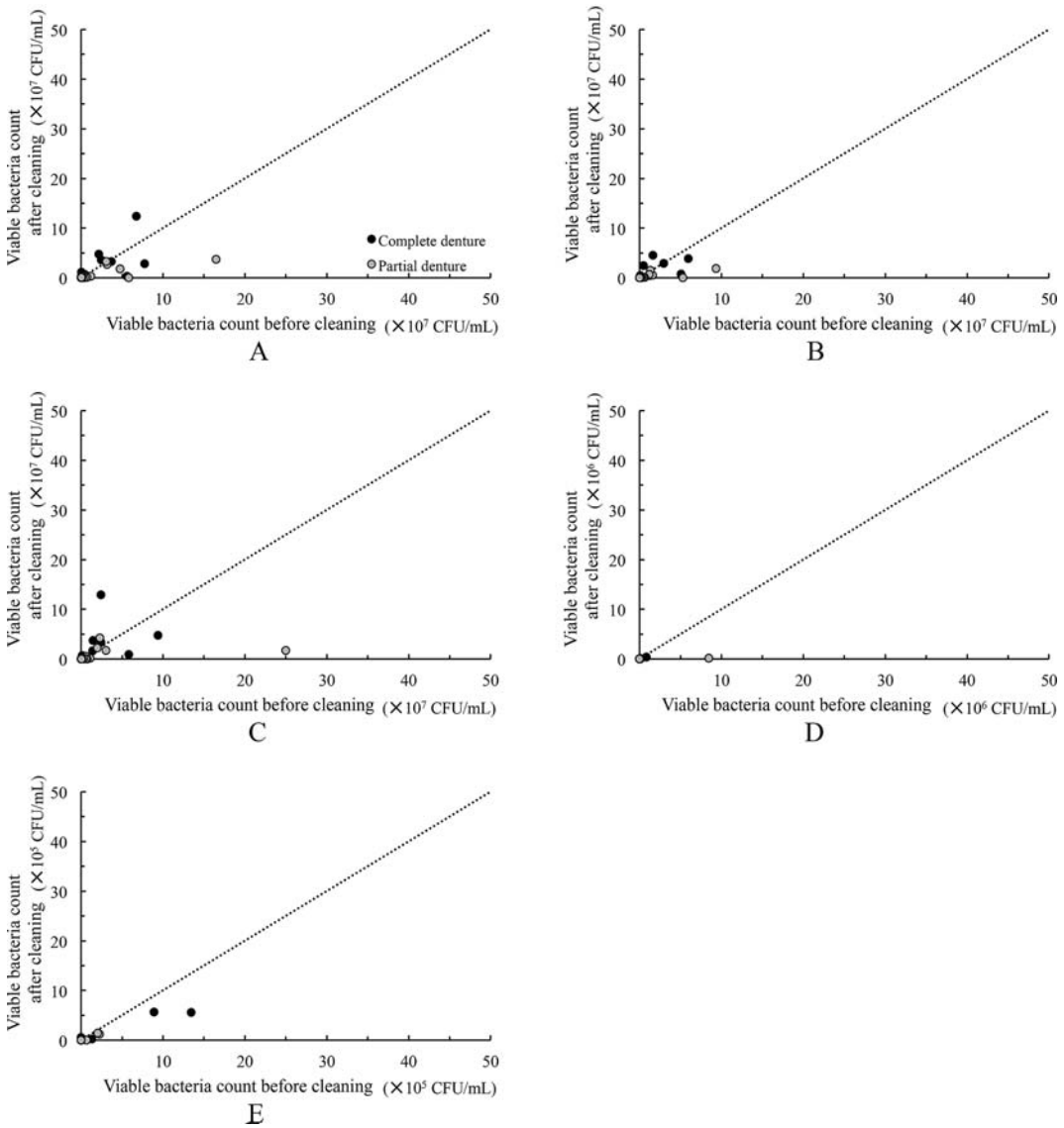

Fig. 9 Relationship between the viable bacteria count before and after cleaning with the combined use of the denture cleaning solution and ultrasonic cleaning for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.* The dotted lines represent change in the viable bacteria count before and after cleaning.

tures before cleaning, and the sterilization rates were similarly low. Based on the results shown in Figs. 9 and 10, the low sterilized bacteria count of aerobic, anaerobic, and streptococci bacteria after the combined use of the denture cleaning solution and the ultrasonic device were found to be related to the low viable bacteria count before cleaning.

DISCUSSION

Numerous studies have been conducted on the sterilization effect of denture cleaners by experimentally forming biofilms on plates.[5, 6] On the other hand, dentures actually used have shown the degradation of acrylic resin due to long-term use,[10] repair, and thickened plaque caused by insufficient cleaning. Therefore, biofilm on dentures in actual

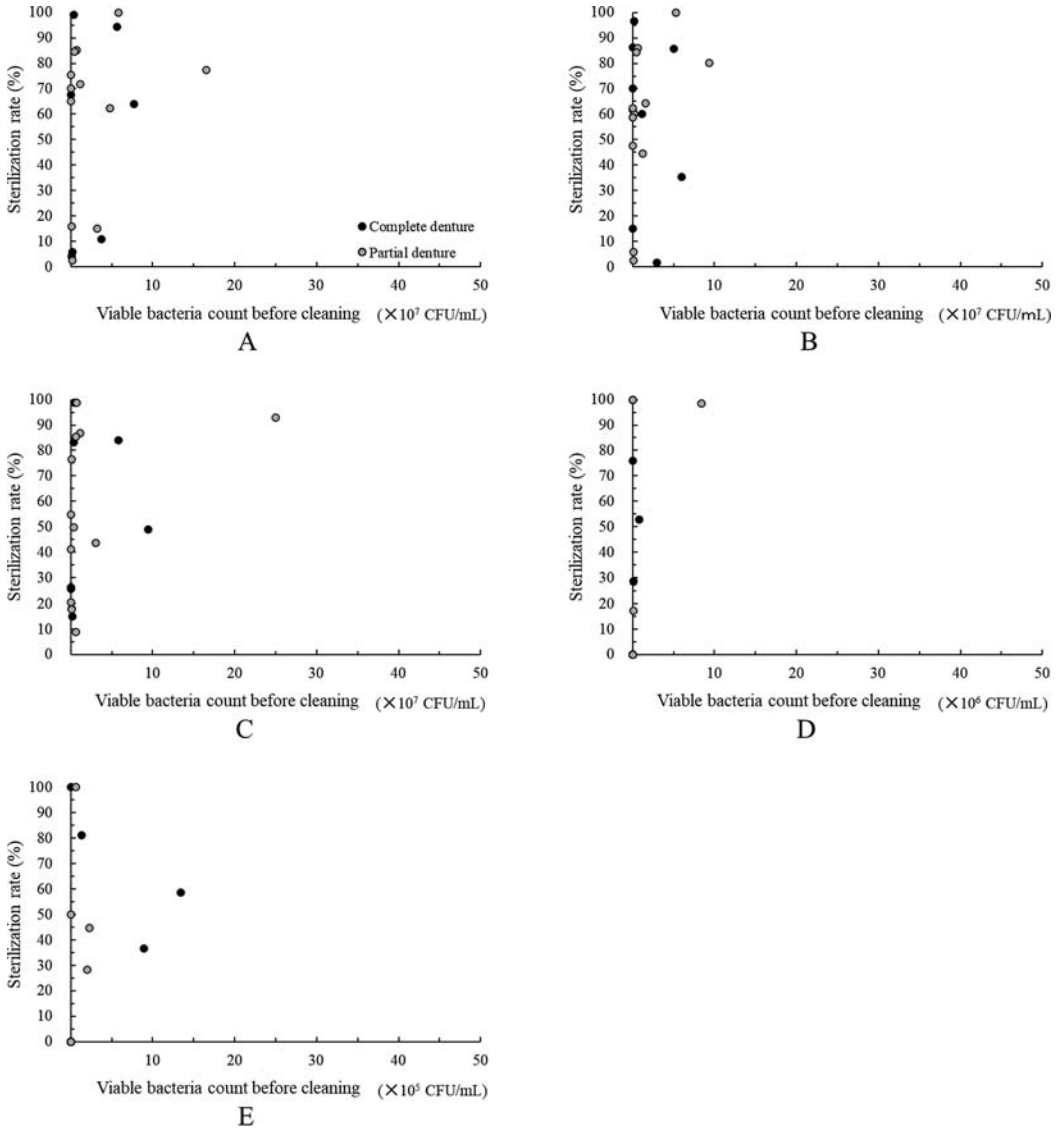

Fig. 10 Relationship between the sterilization rate and viable bacteria count before cleaning with the combined use of the denture cleaning solution and ultrasonic cleaning for (A) *Aerobic bacteria,* (B) *Anaerobic bacteria,* (C) *Streptococci,* (D) *Staphylococci* and (E) *Candida.*

use and that formed artificially on experimental plates are not necessarily the same. Furthermore, the purpose of this study was to examine the actual use of commercial cleaners, not to develop denture cleaners. From this viewpoint, we studied dentures actually used by patients. However, studying dentures actually used by patients introduces problems such as not being able to standardize bacterial counts before cleaning and not being able to collect bacteria from the same location after cleaning because the bacteria count at the place where they were collected before cleaning had decreased. Therefore, in this study, we decided to attempt to reduce the errors caused by these problems and keep the research as symmetrical as possible by collecting bacteria from symmetric sides of the dentures before and after cleaning. In addition, we sought to limit the difference in bacteria counts before cleaning as much as possible by cleaning with a denture brush before immersion in the cleaning

solution.

Nevertheless, as can be seen from Figs. 5, 7 and 9, some dentures had a higher viable bacteria count after cleaning than before. Such a result has been reported in a similar study by Sumi *et al.*[11] The reason for this was thought to be differences in the bacteria collection points on the dentures before and after cleaning, as described above. In other words, it is very difficult to equalize bacteria counts on the left and right sides of dentures actually being used by patients.

Mechanical cleaning before use of denture cleaner

The reason that mechanical cleaning was performed first was not only because we thought that cleaning with a denture cleaner after use of a denture brush would limit the difference in the bacteria count on the dentures before cleaning, as described above, but also because this method is commonly recommended. In this study, saliva and debris on the denture surface were removed by brushing about three times with a denture brush before use of the denture cleaner. On the other hand, as can be seen from the results shown in Figs. 6, 8 and 10, if the bacteria count detected before use of the denture cleaner was low, it was difficult to achieve sterilization. In particular, the results of immersion at 15°C and ultrasonic cleaning showed that the viable bacteria count before cleaning was low, and thus, the sterilized bacteria count and the sterilization rate could not be examined sufficiently. In the future, we would like to consider not performing mechanical cleaning before use of the denture cleaner or prohibiting participants from cleaning their dentures for several days beforehand. It has been said that the deposition of calcium phosphate compounds begins to occur within 3 days.[12] Therefore, we consider that the prohibition period of denture cleaning should be no more than 3 days.

Cleaning conditions

According to the manufacturer's instructions, the effectiveness of both Tough Dent™ and Partial Dent™ decreases at low temperatures. For this reason, it is recommended that it be dissolved in water at 40-50°C. The manufacturer's instructions state that dentures should be immersed overnight to disinfect and bleach them thoroughly, and be soaked for 5 min to remove food debris after a meal. Shirai *et al.*[5] reported that denture cleaners have a significant effect on removing *Candida albicans* 30 min after immersion compared with a control (distilled water); however, it took more than 60 min to remove most of the biofilm-forming *C. albicans.* For Ultrasonic Tough Dent™, the dissolution temperature is less than 40°C, and the cleaning time is basically 5 min.

In this study, considering the dissolution temperature of the denture cleaner and that specified by the manufacturer in combination with ultrasonic cleaning, we set the temperatures at 40°C or 15°C for the denture cleaner, and at 40°C for ultrasonic cleaning. However, because we were keeping dentures that the patients were actually using, the cleaning time with the denture cleaner was set at 30 min, and that with the ultrasonic device at 5 min, as specified by the manufacturer.

Storage of the collected viable bacteria

In this study, the collected viable bacteria were stored in the iSWAB®. In this storage device, live bacteria from samples such as the oral cavity, skin, feces, and soil can be stored at room temperature for up to 8 weeks while maintaining the community composition. Although Inoue *et al.*[7] performed a study similar to ours using physiological saline as the storage solution, in contrast to the results of this study, they reported an extremely high sterilization rate (close to 100%). Therefore, in this study, we stored two cases in physiological saline, and then calculated the number of bacteria removed before and after washing, and the sterilization rate. Both cases showed very high sterilization rates, as in the study by Inoue *et al.*[7] If physiological saline is used as a storage solution, the stored bacteria may be affected in some way, which in turn could affect the sterilized bacteria count and sterilization rate. However, the details underlying this issue remain

somewhat unclear, and thus, should be investigated in a future study.

Influence of denture type and denture cleaner

In this study, in accordance with the manufacturer's instructions, Tough Dent™ and Partial Dent™ were used for the complete and partial dentures, respectively, and a recommended denture cleaner was used with the ultrasonic device. The detailed ingredients of these three denture cleaners were not disclosed. However, the basic components are very similar. Tough Dent™ and Ultrasonic Tough Dent™ are considered neutral cleaners, while Partial Dent™ is considered weakly alkaline, which prevents corrosion of the metal parts. In addition, the special cleaner for the ultrasonic device can be dissolved in a short time because the cleaning time recommended by the manufacturer is short. As shown in Figs. 5-10, the markers for complete and partial dentures were evenly distributed without a significant bias for either the sterilized bacteria count or the sterilization rate. Therefore, hardly any effects were observed in regard to the type of dentures or cleaner. Therefore, in this study, we analyzed the results without separating out these conditions.

Effects of cleaning temperature

Regarding the solution temperature of the cleaner, the sterilization rates of the aerobic and streptococci bacteria were significantly greater at 40°C than at 15°C, as shown in Table 1. As shown in Fig. 6, dentures with a viable bacteria count greater than 4.54×10^9 CFU/mL before rinsing at 40°C showed a high sterilization rate greater than 95%. The manufacturer recommended dissolution in warm water at 40-50°C, since the foaming action weakens when the water temperature is low. In general, because the proteolytic enzymes in denture cleaners show high activity at about 40°C,[13] it is thought that the sterilization effect in a solution is enhanced at 40°C. As for the bleaching activator in the denture cleaner, it is thought that the bleaching effect is enhanced at higher temperatures. Inoue *et al.*[7] also reported that when using denture cleaners, the sterilization is more effective when the water is warm rather than at room temperature. Therefore, it can be said that cleaning in warm water (40-50°C), which can be performed easily at home without affecting the acrylic resin, is effective, as indicated by the manufacturer.

In this study, the temperature of the cleaning solution was examined after immersion at 40°C for 30 min. However, to improve the sterilization effect by immersion for a long time, it is necessary to consider cleaning at 50°C, as the liquid temperature decreases over time. On the other hand, considering that dentures are immersed overnight, a denture cleaner with a high sterilization effect at 15°C needs to be developed.

Combined use with ultrasonic cleaner

In a study by Shiba *et al.*[9] on the effect of a combination of denture cleaner and an ultrasonic device on dentures actually used by patients, the fungal score decreased significantly after 10 minutes of use of the ultrasound compared to before cleaning, but did not decrease significantly until after 20 minutes without ultrasound. It has been suggested that denture cleaning using ultrasound combined with an enzyme-based denture cleaner is effective for maintaining and improving oral hygiene in denture wearers. In the present study, the combined use of an ultrasonic device did not show a strong sterilization effect. This result may be due to the low bacterial count before cleaning with ultrasound. The low viable bacteria count could be observed by comparing the viable counts before cleaning (the horizontal axes in Figs. 6 and 10). In this study, the viable bacteria count of both the aerobic and anaerobic bacteria on dentures cleaned with the combination of ultrasonic cleaning was only about 1/1000 that of dentures cleaned at 40°C. This was thought to be related to the difficulty in unifying the viable bacteria count before cleaning when the dentures actually used by patients were studied as described previously.

Figure 11 shows the results of observations before and after ultrasonic cleaning with warm water at 40°C without using a denture cleaner. The rea-

A B

Fig. 11 Macroscopic comparison (A) before and (B) after cleaning using the ultrasonic cleaner in which the dentures were washed at 40°C in warm water without denture cleaner.

son for showing the results of cleaning without using a denture cleaner is that the cleaning solution becomes turbid, even if the detergent is dissolved. The dentures were considered to have been thoroughly cleaned mechanically with the denture brush ; however, as shown in Fig. 11, after ultrasonic cleaning, the warm water became turbid and adhering substances began floating. Visually, the dentures became very clean after the ultrasonic cleaning, which suggests that even dentures that look clean at first glance still have debris that cannot be removed by mechanical cleaning with a denture brush. However, this debris can be removed by ultrasonic cleaning, which indicates its effectiveness as a mechanical cleaning method. In other words, although no effective sterilization results were observed in this study, it is thought that sufficient debris removal can be expected.

In the elderly, the mechanical cleaning of dentures with a denture brush becomes difficult because of a decreased ability to carry out activities of daily living and an increased need for caregiver support. It is therefore thought that the use of denture cleaners and ultrasonic cleaners in the elderly is becoming increasingly important. In a future study, we would like to consider the effects of the combined use of ultrasonic cleaning from the viewpoints of both the removal of deposits by mechanical cleaning and the disinfection effect of chemical cleaning. At the same time, we would like to study this in more detail by measuring the bacteria count in dentures with a high viable bacteria count before cleaning, or by measuring the suspended bacteria in the cleaning solution after denture cleaning.

Evaluation of cleaning effects

The sterilization rate is described on packaging as an indicator of the effect of commercially available denture cleaners. However, as shown in Figs. 6, 8 and 10, as an index, the sterilization rate is greatly affected by the viable bacteria count before cleaning. In other words, the sterilization rate increases with an increase in the viable bacteria count before washing, while it decreases with a decrease in the count. In this study, the viable bacteria count after cleaning was greater than that before cleaning, and several dentures had a negative sterilization rate. On the other hand, as can be seen in Fig. 5, the viable bacteria count is suppressed to a certain level after use of the denture cleaner, regardless of the count before cleaning. Therefore, in dentures actually used by patients, since the sterilization rate varies greatly depending on the viable bacteria count adhering to the denture, it may be necessary to consider evaluating denture cleaners based on the viable bacteria count adhering to the denture as opposed to the sterilization rate.

Countless resident bacteria can be found in the oral cavity. Even if the dentures can be completely sterilized, they will be contaminated with resident bacteria as soon as they are worn in the mouth. From this point of view, it is necessary to consider

not only how to sterilize bacteria from dentures completely when cleaning them, but also how much the viable bacteria must be sterilized when adhered. In addition, the bacteria count remaining after washing is considered to affect not only the sterilizing power of the denture cleaner, but also the properties of the dentures themselves. Nakazato *et al.*[14] reported that there was no significant difference in bacterial counts or species among various materials for the denture base in the oral cavity. On the other hand, they stated in the same paper that dentists also need to take care to minimize plaque adhesion by polishing the base after adjusting the dentures. In conclusion, in the future, to evaluate the sterilization effect of denture cleaners on dentures actually used by patients, it is necessary to consider the viable bacteria count that adheres to the denture after cleaning, the cleaning and sterilizing effects of the denture cleaner, and the shape, material and surface roughness of the prosthesis.

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