The Relationship between the Physical Properties of Oral Moisturizer and the Denture Retention Force

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Abstract

Purpose The purpose of this study is to determine whether an oral moisturizer can serve as a denture adhesive and to clarify the effects of different types of oral moisturizers and their physical properties on denture retention force.

Method Twenty-one types of oral moisturizer and two types of denture adhesive as controls were used as test materials. Spinnability tests, viscosity tests, and the evaluation of retention strength were carried out. A statistical comparison of the test results was performed using one-way ANOVA and a multiple-comparison test.

Results Mean viscosity was 1.5×10^2 mP·s for the spray-type oral moisturizer was mPa·s, 3.0×10^3 mPa·s for the liquid-type, 1.5×10^5 mPa·s for the gel type, and 1.4×10^6 mPa·s for the denture adhesive.

Mean retention strength was 3.6 N for the spray-type oral moisturizer, 14.4 N for the liquid-type, 30.1 N for the gel-type, and 36.0 N for the denture adhesive. There was a positive correlation between the viscosity and the retention strength of the oral moisturizers ($r = 0.98$, $p < 0.01$).

Discussion The gel-type oral moisturizers had the same level of viscosity and provided the same retention strength as the denture adhesives used as controls, suggesting that a gel-type oral moisturizer may serve as a denture adhesive.

Keywords. denture retention, oral moisturizer, spinnability, viscosity,

Introduction

Hyposalivation is a disorder of the salivary gland that can occur after surgery for oral cancer. Dentures may be used to compensate for the structural and functional loss due to oral cancer surgery. However, the use of dentures can be difficult on hyposalivation. Patients with complete dentures and oral dryness suffer from various problems including frequent oral ulcers, delayed healing of the oral membrane, and reduction of the retention force of complete dentures.

Therefore, we examined whether oral moisturizers, which are often used to treat oral dryness and to increase the retention force of dentures, can be used as substitutes for denture adhesives. Most oral moisturizers contain salivary or antibacterial components, as well as components that promote the maintenance of moisture and humectants in the oral cavity. They are widely recommended to patients with oral dryness and for people who need nursing care. However, commercially available oral moisturizers have different physical properties, medicinal effects, smells, and tastes; consequently, a suitable oral moisturizer should be carefully prescribed for each particular case.

To assist in the selection of appropriate oral moisturizers, we sought to clarify the effects of the different physical properties of various oral moisturizers on denture retention force, in order to examine whether oral moisturizers can be used as substitutes for denture adhesives. We believe that, in the future, the retention force of complete dentures may be improved by selecting an oral moisturizer on basis of the degree of oral dryness.

Methods

1. Test specimens

In this study, 27 test specimens were used (Table 1): 21 oral moisturizers (4 sprays, 4 liquids, and 13 gels), 3 denture adhesives, 1 artificial saliva, and 3 imitation saliva solutions (glycerin solution).

2. Variables measured

We measured the spinnability and viscosity of the various oral mois-

Table 1. Test specimens

Type	CODE	Trade Name	Manufacturer	Address
Artificial saliva	a	Saliveht	Teijin Pharma	Tokyo
Imitation Natural Saliva Solution	b	Concentrated Glycerin 85 %	Dynamo	Osaka
	c	Concentrated Glycerin 70 %		
	d	Concentrated Glycerin 60 %		
Spray	A	Wet Care	Kissei Pharma Ceutical	Nagano
	B	Oral Refre	Dia - Medical Supply Morita	Tokyo
	C	Oral Wet Spray	Yoshida	Tokyo
	D	Stoppers for	Sun Dental	Tokyo
Liquid	E	Mouth Moist Plus	Mind Up	Yamanashi
	F	Fit Angel	Panasonic Dental	Tokyo
	G	Aqua Mucus Liquid	Life	Saitama
	H	Oral balance Liquid	T&K	Tokyo
Gel	I	Ubra Gel	Bee Brand Medico Dental	Tokyo
	J	Honey Wet	Nippon Zettoc Morita	Tokyo
	K	Oral Refre Jell	Ryoka Dental Morita	Ibaraki
	L	Viva - Jellwet	Tokyo giken	Tokyo
	M	Refre-care H	En Otsuka Pharmaceutical	Tokyo
	N	Wet Keeping	Oral Care	Tokyo
	O	Biotene Oral balance Jell	T&K	Tokyo
	P	Fit Angel Jell	Panasonic Dental	Tokyo
	Q	Bioxtra Aqua Mouth Jell	Weltec	Osaka
	R	Aqua Mucus Gel	Life	Saitama
	S	Oral Aquagel	Gc	Tokyo
	T	Denture Gel	Kamemizu Chem.Ind	Osaka
	U	Mouthpure	Kawamoto - Sangyo. Co., Ltd.	Osaka
Denture Adhesive	V	Poli • Grip	Earth Chemical Co., Ltd.	Tokyo
	W	Mizugrip	Kobayashi Pharmaceutical Co., Ltd.	Osaka

turizers. We also measured their retention force using an experimental denture base to evaluate their adsorptive power.

1) Measuring spinnability

Spinnability was measured using a Neva Meter® (Technical Solutions Co ISHIKAWA IRON WORKS, Kitakyushu): 0.06 mL of test specimen was measured at a rate of 0.50 mm/s in dry mode. Spinnability was obtained by performing seven measurements for each specimen and taking the mean of five of the measured values (excluding the maximum and minimum values).

2) Measuring viscosity

Viscosity was measured using the standard test method: a digital rotational viscometer (Brookfield rotational viscometer, Osaka) was used to measure two 500 mL samples of each test specimen at a temperature of 20℃. The two measured values for each specimen were averaged and the mean value was taken as the viscosity of the specimen.

3. Measuring retention force

1) Developing a retention force measurement device

The experimental denture base section of the retention force measuring device was prepared by taking an impression (Examixfine® injection type) of an edentulous upper jaw model (G10 FE - 402 K, Nissin, Tokyo). Five experimental denture bases were produced using a room-temperature curing resin and an upper edentulous model. An experimental denture formed a bite plate and the denture base mucosal surface was relined to compensate for polymerization shrinkage (Fit Resin®, Shofu, Kyoto). In addition, a 5.0 mm diameter ring was fixed vertically on the center of the denture base.

2) Traction method

A suitable amount of each specimen was dropped or otherwise applied between the model and the experimental denture base and a load of 2.5 kg was applied using a disc weight. The load was applied vertically to the denture base at a rate of 0.5 N/s and it was measured using a push-pull gauge. Three measurements were performed for each specimen on five experimental denture bases.

4. Statistical analysis

The significance of differences was analyzed by one-way ANOVA or a multiple comparison test. A significant relationship was determined based on the Pearson's correlation coefficient (r). Relationships yielding p-values of less than 0.01 were considered significant.

The statistical software SPSS (SPSS 14.0-J for Windows) was used in the statistical analysis.

Results

1. Spinnability

Figure 1 shows the results for the spinnability measurements. The artificial and natural imitation salivas, as well as the spray and liquid oral moisturizers, exhibited very similar average spinnabilities in the approximate range 3–4 mm. By contrast, the gel oral moisturizers had spinnabilities over a wide range from 2 to 23 mm. The two denture adhesives had similar spinnabilities to the other test specimens (excluding the gel oral moisturizers). The spinnability test results revealed no statistically significant differences among the groups.

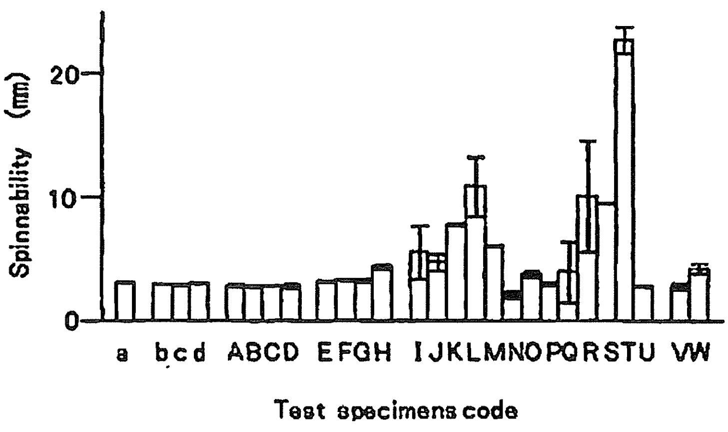

Figure 1. Measuring spinnability.

2. Viscosity

Figure 2 shows the viscosity measurements. The measured viscosities ranged widely from less than 10 to greater than 10^6 mPa·s. As Figure 2 demonstrates, the artificial saliva, imitation saliva solutions, and spray oral moisturizers all had very similar viscosities, whereas the liquid oral moisturizers were slightly more viscous. The gels and the denture adhesives had very similar viscosities that were considerably higher than those of the liquid oral moisturizers. The viscosity of the denture adhesive Poligrip® could not be measured because it exceeded the measurement range of the measuring device. The viscosities of the gels and the denture adhesives differed significantly from those of the other test specimens.

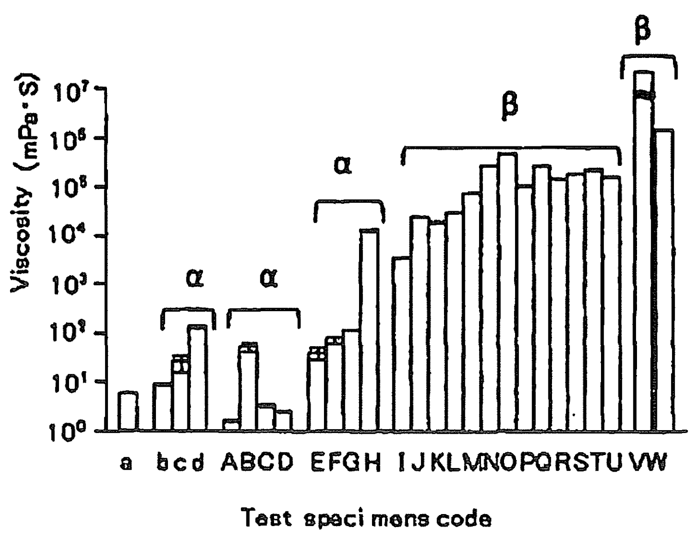

Figure 2. Measuring viscosity. Different Greek letters respresent significant differences($p < 0.05$, $\alpha < \beta$).

3. Retention force

Figure 3 shows the results of the retention force measurements. The artificial saliva, imitation saliva solutions, and the spray moisturizers gave very similar retention forces. The retention forces of the liquid moisturizers differed significantly from those of the sprays, artificial saliva, and natural imitation saliva solutions. The gels and denture adhesives had significantly higher retention forces than the liquid oral moisturizers.

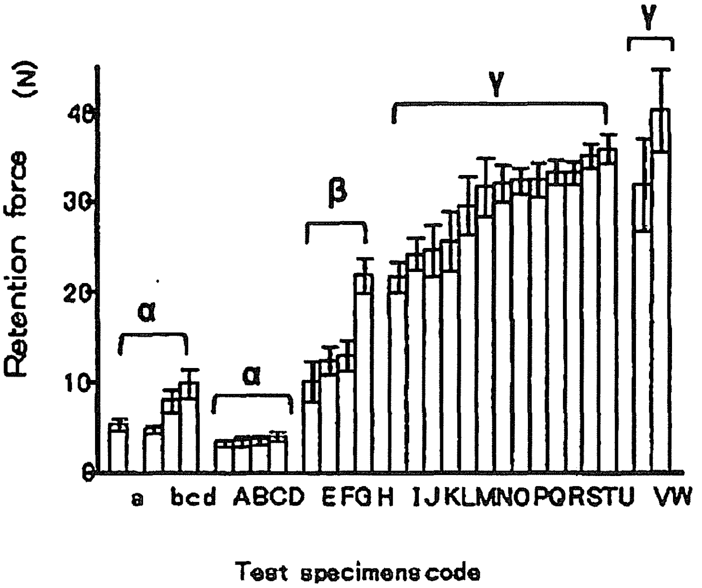

Figure 3. Measuring retention force. Different Greek letter represent significant differences ($p < 0.05$, $\alpha < \beta < \gamma$)

4. Relationship between spinnability and viscosity

The spray oral moisturizers, imitation saliva solutions, artificial saliva, and the liquid oral moisturizers had very similar spinnabilities and viscosities that were rather low. By contrast, both the spinnabilities and the viscosities of the gel oral moisturizers varied widely between products. No significant correlation was found between the spinnability and the viscosity of the test specimens [$r = 0.42$, $p = 0.96$ (>0.01)].

5. Relationship between spinnability and retention force

The gel oral moisturizers exhibited high retention forces, but their spinnabilities varied widely between products. The other test specimens had very similar spinnabilities, whereas their retention forces varied widely between specimens. No significant relationship was found between the spinnability and the retention force of test specimens [$r = 0.41$, $p = 0.11$ (>0.01)].

6. Relationship between retention force and viscosity

Figure 4 shows the relationship between the viscosity and the retention force: the retention force increases as the viscosity increases. These two parameters display a statistically significant, and positive correlation between them ($r = 0.98$, $p = 0.01$).

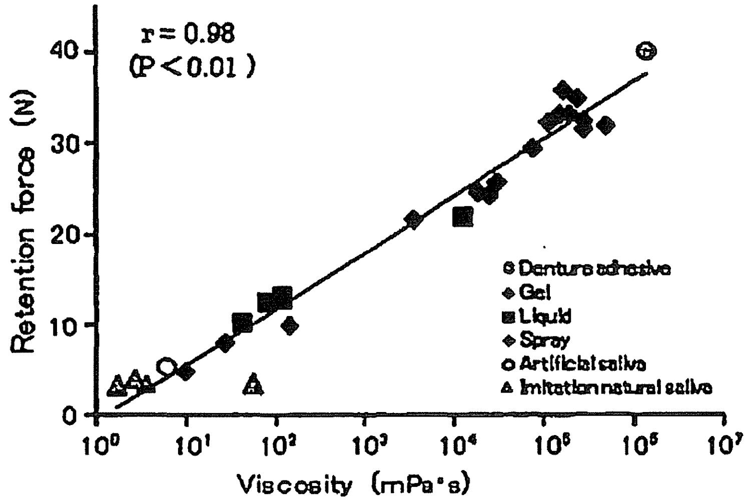

Figure 4. The relationship between viscosity and retention force.

Discussion

I. Methods

1. Selection of test specimens

In this study, we used 21 currently available commercial oral moisturizers. In addition, two denture adhesives (creams), an artificial saliva, and three imitation saliva solutions were used as controls. We selected three types of moisturizer (sprays, liquids, and gels). Each oral moisturizer was manufactured according to the manufacturers' standards to satisfy specifications for viscosity, pH, specific gravity, and degree of foaming. However, the details of their manufacturing specifications are not publicly available. Consequently, we first compared the physical properties of the three types of oral moisturizers and their effect on denture retention.

2. Selection of spinnability measurement device

Spinnability is a rheological property of saliva. It is usually measured from a small specimen using a Neva Meter®, an instrument that measures the physical properties of materials including saliva and cream. It is compact and lightweight, enabling the measurement of fluids such as saliva at the chair side (Kakinoki Y, 2002). Spinnability was one of the variables tested in the study of the physical properties of saliva reported by Kakinoki (Kakinoki Y, 2003). In this study, we performed measurements in the dry mode because many of the test specimens were susceptible to plastic deformation.

3. Selection of viscosity measurement device

We used a digital rotational viscometer to measure viscosity. This device is suitable for measuring non-Newtonian viscosity, a rheological property of highly viscous materials. A digital rotational viscometer can also be used to measure various physical properties of low- and high-viscosity liquids by simply changing the adapter and rotor.

4. Selection of denture retention measurement device

In this study, we used a model to determine the differences in physical properties and retention forces of different kinds of oral moisturizers when tested under the same conditions. The experimental denture base was prepared in the form of a bite plate in order that a load could be applied to it and it could then be pulled, simulating the conditions in the oral cavity.

On the basis of the study by Bandai M et al, a load was applied for 10 s before measuring the retention force. The weight and traction speed were set to 2.5 kg and 0.5 N/s, respectively, since these settings were found to give stable retention force readings in a preliminary experiment. The amount test specimen to be used was determined in the preliminary experiment, which revealed that using an amount of test specimen that just covered the whole surface of the model resulted in stable retention force measurements.

Ⅱ. Results

1. Spinnability

Spinnability depends on the stringiness length and the viscosity. The seven gels had spinnabilities of 5 mm or greater. The spinnabilities of the other test specimens mostly lay in the spinnability range for the saliva of a normal person at rest (2–5 mm), as reported by Kakinoki (Kakinoki Y, 2003). The high spinnabilities of the gels are considered to be because of their high viscosities, stagnation properties, and fluidities. They are expected to have a high moisturizing effect. The spinnability of a material depends on its viscosity, surface tension, and elasticity coefficient. In particular, a high viscosity is related to a low spinnability. Denture adhesive feels like a gel or a cream and it readily undergoes plastic deformation. For this reason, the denture adhesives used in this study had high viscosities and low spinnabilities.

2. Viscosity

The viscosities were measured in previous studies for saliva or natural imitation saliva solutions (Bandai M et al., 1987; Kakudo Y 1976; Katayama S et al., 1985; Kawanuma Y 1982; Komatsu S et al., 1991; Takahashi M, 1998). In this study, the artificial saliva had a viscosity of approximately 6.0 mPa·s, which is within the range specified by the manufacturer (i.e., 4–6 m^2/s at a temperature of 25℃). The viscosities of the imitation saliva solutions were quite different from those reported in the studies mentioned above.

Both Bandai and Takahashi reported the same viscosities of the imitation saliva solutions as we obtained in our study. However the glycerin contents of the imitating saliva solutions differed between our study and studies of Bandai and Takahashi. We believe that this difference in glycerin content is due to difference in temperature.

Because many of the products used in this study, such as oral moisturizers and denture adhesives, are usually stored at room temperature, the viscosity measurements were performed at room temperature (i.e., 20℃). Therefore, a temperature difference is the most probable cause for the large difference in the viscosities obtained by the present study. In addition, the high glycerin contents of the imitation saliva solutions gave high viscosities. Many oral moisturizers contain glycerin solution for moistening and wetting. The glycerin solution has a high viscosity and stagnation in the oral cavity. However, excessively high glycerin content has been reported to give rise to a high hygroscopicity. Thus, high-viscosity oral moisturizers may aggravate oral dryness.

The possibility that measurement temperature and glycerin content would affect viscosity was suggested. These results reveal that the temperature at which the measurements are obtained and glycerin content may influence viscosity.

3. Retention force

Many studies have investigated measurements of denture retention force (Bandai M et al., 1989; Cumhur S, 2007; Kakudo Y, 1976; (Sato Y et al., 2009; Sekine H et al., 1963). Oral cavity retention force often appears to be affected by the amount and quality of a person's saliva and the form of an individual's alveolar ridge. In order to measure and compare the retention forces of oral moisturizers under the same conditions, this study included an experiment performed using a model. Seven of the gel moisturizers investigated in this study exhibited higher retention forces than the denture adhesive Poligrip®. These results suggest the possibility of substituting a gel for denture adhesive.

4. Relationship between spinnability and viscosity

The gel-type oral moisturizers were classified into three types according to their physical properties. Their applications may vary depending on their physical properties.

The first type exhibited a high spinnability and a low viscosity. This type of moisturizer has good properties over the oral cavity but it does not stagnate there. Therefore, this type may alleviate oral dryness temporarily in common with sprays and liquids.

The second type exhibited a high spinnability and a viscosity ranging from low to high. Well-fitting dentures have an appropriate border seal between residual ridge tissue and the denture base. The border seal increases the retention force of a complete denture. We hypothesize that spinnability is a property related to spread within the oral cavity and that viscosity is a property related to stagnation. It is possible that certain of the oral moisturizers that are used as saliva substitutes, such as the sprays, liquids and this type of gel moisturizer, which spread thinly over the whole denture, can increase the retention force of the complete denture. With this type it may be possible to select moisturizers of different viscosities according to the grade of mouth dryness. We believe that with this type of moisturizer it may be possible to increase the retention force of a complete denture in a patient secreting low amounts of saliva.

Conversely, a large space between residual ridge tissue and the denture base is caused by ill-fitting dentures. If this large space can be decreased, we believe that the retention force of a complete denture may be increased. Therefore, we propose that an oral moisturizer with properties similar to denture adhesives will increase the retention force of the complete denture.

The third type of moisturizer exhibited similar properties to denture adhesive. This type showed low spinnabilities and viscosities ranging from moderate to high. This type may have longer stagnation times in the oral cavity than the other two types and may, therefore, be a suitable substitute for denture adhesive. We believe that it is possible to increase the retention force of a complete denture by utilizing this type of moisturizer.

5. Relationship between spinnability and retention force

Many studies have investigated factors that relate to the retention force of dentures. However, no study has investigated the relationship between spinnability and retention force. Therefore, we examined the relationship between spinnability, a rheological property of saliva, and the denture retention force of various specimens. The results revealed that there was no correlation between spinnability and retention force. However, three of the high-spinnability gels gave a higher retention force than the denture adhesive Poligrip®. These specimens may increase the retention force of dentures by forming a thin layer between the dentures and the mouth mucosa. Four of the low-spinnability gels gave a higher retention force than Poligrip®. These gels have good properties in the oral cavity. Moreover, they may have similar physical properties to Poligrip®. Therefore, they may increase the retention force of dentures by stagnation in the oral cavity.

6. Relationship between retention force and viscosity

Many studies have investigated the relationship between denture retention force and the viscosity of liquids (Bandai M et al., 1987; Nagao M et al., 1982; Takahashi M, 1998. However, these studies used a small number of test specimens that had similar characteristics. We used 27 oral moisturizers that had different physical properties and we found a positive correlation between viscosity and retention force.

A complete denture requires three physical properties: static adhesion, dynamic adhesion, and adhesive power (Yamagata K et al., 2004). It is notable that, in our study, test specimens with a high viscosity were resistant just before the complete denture broke away. Therefore, we believe that it is possible for high-viscosity oral moisturizers to display an equivalent retention force to denture adhesive within the oral cavity. However, it is necessary to study the stagnation and transpiration of oral moisturizers in the mouth.

In the present study, glycerin solutions diluted to 60, 70, and 85% had viscosities of 9.7, 27.0, and 140 mPa·s, respectively. According to the results of Takahashi (Takahashi M, 1998), these three solutions had average denture retention forces of 4.8, 8.0, and 9.9 N, respectively. Compared with these results in our study, the minimum denture retention force was very similar but the maximum denture retention force was smaller. We believe that the difference in our result may have been caused by the temperature at the time of measurement and the method of measurement used.

In this study, the artificial saliva and the spray and liquid oral moisturizers displayed denture retention forces similar to those previously reported for glycerin solutions and saliva. By contrast, gel oral moisturizers showed higher viscosities and denture retention forces. This result suggests that oral moisturizers may increase the denture retention force of denture wearers with oral dryness. In addition, oral moisturizers with high viscosities exhibited an equivalent denture retention force to denture adhesive. We intend to examine the transpiration and stagnation properties of oral moisturizers in future studies. Furthermore, the positive correlation between viscosity and denture retention force suggests that viscosity contributed largely to the high denture retention forces of high-viscosity oral moisturizers.

Conclusion

The denture retention force of oral moisturizers was related to viscosity, whereas it was not related to spinnability. There was no correlation between the viscosity and the spinnability. However, we were able to classify gel-based oral moisturizers into three types on the basis of their retention force, viscosity, and spinnability; these types may be used in different ways for treating the symptoms of oral dryness or denture retention. The denture retention force of oral moisturizers was related to their viscosity. High-viscosity oral moisturizers had a similar denture retention force to those of denture adhesives. This suggests that high-viscosity oral moisturizers may be suitable as substitutes for denture adhesives.

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Gerodontology

Original article

Factors affecting the formation of membranous substances in the palates of elderly persons requiring nursing care

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Factors affecting the formation of membranous substances in the palates of elderly persons requiring nursing care

Objective: To determine the causative factor behind the formation of membranous substances in the mouths of elderly patients requiring nursing care.
Background: Membranous substances are sometimes observed in the mouths of elderly persons requiring nursing care, and these can lead to bleeding, infection and asphyxiation.
Materials and methods: In April 2007, samples were collected from 70 patients at C Hospital, Aichi Prefecture, Japan, who were 65 years or older (median age, 81.1 ± 7.7 years). Sixteen of the subjects were confirmed to have a membranous substance containing a keratin degeneration product that had been derived from stratified squamous epithelium. The samples were examined microscopically, and the presence of epithelial components was confirmed through immunohistochemical staining with anti-cytokeratin-1 antibodies.
Results: Decision tree analysis and logistic regression suggest that the leading contributors to the formation of the membranous substances were the method of ingesting nutrients, dryness of the tongue dorsum and open mouth. These three factors are related to elderly persons requiring nursing care with impaired oral cavity function, and it was suggested that dryness of the oral mucosa was the major factor behind the membrane formation.

Keywords: peeling epithelial membrane, elderly patient, oral hygiene, dry mouth

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Introduction

Oral care, such as assisted tooth brushing, is essential for maintaining the oral health of elderly persons requiring nursing care. Furthermore, in elderly patients with serious conditions such as impaired consciousness, oral care contributes to the maintenance of systemic health, for example, in the prevention of aspiration pneumonia[1]. A general rule of thumb is that the more serious the case, the more important oral care for elderly persons requiring nursing care becomes. However, membranous substances, which are not observed in healthy individuals, are sometimes observed in the mouths of elderly persons requiring nursing care. These are often reported to be in the form of a crust[2,3] or sputum[4–7], but there are reports suggesting that they are actually a peeling epithelial membrane. This latter description is based on the fact that epithelial components exhibiting a layered structure with corneum have been seen upon pathological observation of these membranes[8,9]. Some of these membranes are dry, and some are viscous. They may be removed using tweezers, as a lump, but some of the substance occasionally gets left behind in the oral mucosa. Membranous substances that adhere to the palate not only affect speech when they fall down the throat, but they are also believed to cause danger from suffocation. However, it is not clear what types of patients are prone to the formation of membranous substances inside the mouth. Clarifying the formative mechanisms of these membranous substances is believed to be the key in preventing the formation of these membranes, which will improve the quality of life for terminal elderly persons requiring assisted dental care.

In this study, membrane-like substances sampled from elderly patients in a nursing care facility were microscopically observed, the presence of epithelial components was confirmed and the factors affecting the formation of the membranes were investigated. Multivariate analysis was used for the investigation of related factors, and a decision tree analysis was used to show the interrelationship between the independent variables (explanatory variables) and the dependent variables involved in the formation of peeling epithelial membranes. Multivariate logistic regression was used to investigate the level of factor correlation.

Subjects and method

Research subjects

The subjects were 70 elderly patients requiring nursing care in C hospital, Aichi Prefecture, Japan, and samples were collected in April 2007. The patients were 65 years or older (median age, 81.1 ± 7.7 years) and consisted of 24 men and 46 women. All study subjects were bedridden, and assisted tooth brushing was carried out twice a day. This study was approved by the ethics committee of Matsumoto Dentistry University (approval number 39), and approval was given by all patients following sufficient explanation to family members regarding this study.

Method

From the hospital records, the age, diseases, commonly used drugs, degree of bedriddenness, level of consciousness and ability to communicate were determined for each of the patients. Information on the frequency of assisted tooth brushing was obtained orally from the attending nurse. The dentist used a flashlight and mirrors to evaluate the presence of membranous substance, the gingival index, the presence of an open mouth and tongue coating (according to the classification scheme of Kojima[10], Table 1). Membranous substances that had formed on the palate were removed as much as possible by the dentist using tweezers. The degree of moisture retention of the tongue dorsum (the mucous membrane below the tongue) was evaluated using a 10-s testing method with a wetness tester[11] (KisoWeT®, KISO Science Co., Ltd., Yokohama City, Kanagawa Prefecture, Japan, Fig. 1).

Table 1 Tongue coating classification (Kojima Classification)

Index 1	Thin coating on less than 1/3 of the back of the tongue
Index 2	Thin coating on less than 2/3 of the tongue, or thick coating on less than 1/3 of the tongue
Index 3	Thin coating on more than 2/3 of the tongue, or thick coating on less than 2/3 of the tongue
Index 4	Thick coating on more than 2/3 of the tongue

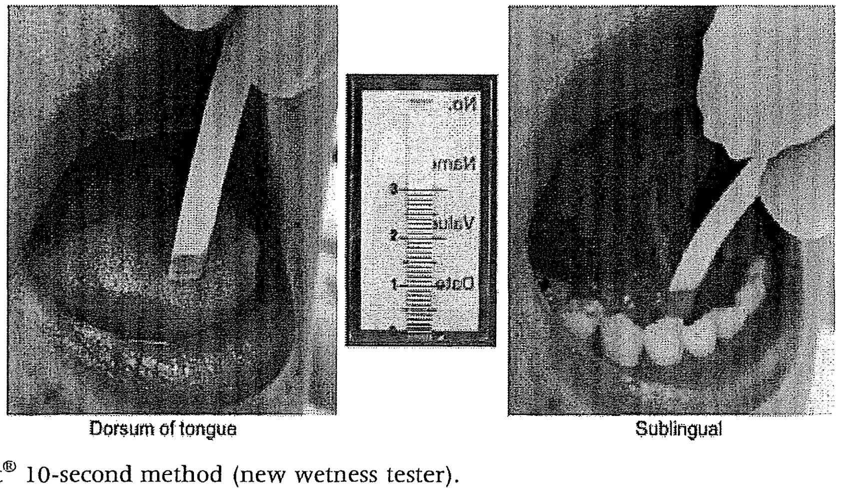

Figure 1 Kiso Wet® 10-second method (new wetness tester).

Membranous substances sampled from the palates of the patients were fixed with a 10% neutral-buffered formalin solution, and HE staining of the paraffin section was carried out using general procedures. For immunostaining of the paraffin sections with anti-cytokeratin-1 antibodies, the samples were autoclaved in a 0.01 M citrate buffer solution (pH 6.0) for 15 min at 121°C according to general procedures[12]. To confirm the presence of layered epithelial structures in the samples, mouse monoclonal anti-cytokeratin-1 antibodies (1:20 dilution, clone 34βB4; Novocastra Laboratories Ltd., Newcastle, UK) were incubated with the samples for 12 h at 4°C[13]. Secondary antibodies were incubated with the samples for 30 min at room temperature using Histofine Simple Stain MAX PO (Multi; Nichirei Co., Tokyo, Japan). Following colouring with 3-3′-diaminobenzidine tetrahydrochloride (Dako, Glostrup, Denmark)[14], the samples were counterstained with haematoxylin. Samples that were confirmed to have keratin degradation products derived from stratified squamous epithelium were classified as membranous substances with epithelial components[8]. Furthermore, Periodic Acid-Schiff (PAS) and mucicarmine staining were performed for other components, and proteins derived from mucin that showed positive reaction to both were analysed.

Statistical analysis

A decision tree analysis was carried out with confirmation of the presence of membranous substances as the dependent variable and a total of 61 independent variables: 14 variables from the patients' general and oral health histories, 15 disease variables and 32 variables for commonly used drugs. The resulting tree diagram provided a visual representation of the relationship between these input variables and their relative impact on the formation of membranous substances. The decision trees were calculated using the chi-squared automatic interaction detection (CHAID) method[15,16]. The entire sample of independent variables (parent nodes) was divided into two child nodes (subgroups) based on the independent variable with the highest chi-squared value in relation to the dependent variable. This procedure was repeated, and the analysis was terminated when the p value was above 5% for any combination of membranous substances as dependent variable and the 61 independent variables. Logistic regression was carried out to obtain an adjusted odds ratio with factors extracted from the decision tree analysis as the independent variables and the formation of a peeling epithelial membrane as the dependent variable. Decision tree analysis was carried out with SPSS Decision Tree (IBM Japan Co., Ltd., Tokyo, Japan), and logistic regression was carried out with SPSS Base System 15.0 (IBM).

Results

Pathological findings

Membranous substances were sampled from the palates of 16 of the 70 research subjects (Fig. 2). The membranous substances were composed of eosinophilic layers interspersed with structureless substances that faintly stained with haematoxylin in some areas. Some samples were accompanied by a small amount of inflammatory cells and/or bacterial mass with the same layered structure as the main constituents of the membranous substance. The eosinophilic layers were cytokeratin-1-positive, so the sampled membranous substances contained keratin degradation products derived from stratified squamous epithelium (Fig. 3).

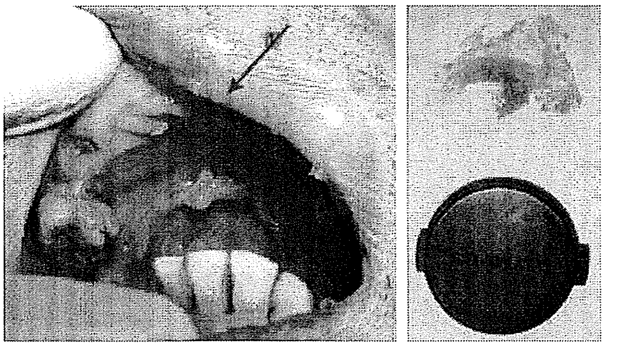

Figure 2 Membranous substance in the palate.

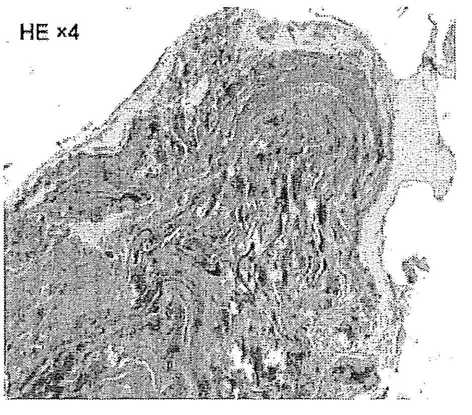

Figure 3 Histopathological observation of a membranous material sampled from the palate of a patient.

Independent correlation analysis

All sampled membranous substances were found to be peeling epithelial membranes, and seven independent variables from the patients' medical histories, including the degree of bedriddenness ($p < 0.001$), the degree of understanding ($p = 0.01$), speech ($p < 0.001$), communication ($p < 0.001$), oral ingestion/intubation ($p < 0.001$), open mouth ($p < 0.001$) and wettability of the tongue dorsum ($p < 0.001$), were found to be correlated with formation of the peeling epithelial membranes (Table 2). Four of the fifteen diseases tested, including dementia ($p = 0.003$), gastrointestinal disease ($p = 0.007$), bone and joint disease ($p = 0.004$) and skin disease ($p = 0.001$, Table 3), and four medications, including lactobacillus preparations ($p < 0.001$), expectorants ($p = 0.002$), antibacterial agents ($p < 0.001$) and lactose ($p = 0.035$, Table 4), were also found to be independently correlated with formation of the membranous substances.

Formative factor determination

A decision tree analysis was carried out with all 70 cases (node 0) as the parent node (Fig. 4). Node 0 consisted of patients who were positive for formation of the peeling epithelial membrane (22.9%) and patients who did not have the membranes (77.1%). Upon analysis, oral ingestion/intubation (the method for ingesting nutrients),

Table 2 Correlation between patient's background, oral findings and exfoliated epithelial membranes ($n = 70$)

		Exfoliated epithelial membrane			
Item	*Category*	*No*	*Yes*	*Total*	*p Value*
Sex	Male	17	7	24	0.383
	Female	37	9	46	
Age	Over 65 years old	10	4	14	0.896
	Over 70 years old	24	6	30	
	Over 80 years old	18	5	23	
	Over 90 years old	2	1	3	
Bedridden level	B Rank	29	0	29	<0.001
	C Rank	25	16	41	
Consciousness level	Awake	47	12	59	0.259
	Awake with stimulus	7	4	11	
Oxygen therapy	Yes	1	1	2	0.407
	No	53	15	68	
Intelligibility	Cannot understand	20	12	32	0.010
	Sometimes able to understand	34	4	38	
Speech	Yes	30	0	30	<0.001
	No	24	16	40	
Communication	Unable	24	16	40	<0.001
	Able	30	0	30	
Oral × Intubation	Ingestion	30	0	30	<0.001
	Tubal feeding	24	16	40	
Aperture	Constant aperture	7	15	22	<0.001
	Close	47	1	48	
Sublingual moistening level	0 mm	8	3	11	0.705
	> 0 mm	46	13	59	
Dorsum linguae moistening level	0 mm	3	14	17	<0.001
	> 0 mm	51	2	53	
Coated tongue	No	35	11	46	0.506
	<30% of dorsum linguae	13	2	15	
	30% or more of dorsum linguae	6	3	9	
Gingival index	Edentulous jaw	20	4	24	0.173
	0	6	0	6	
	Over 0.1	28	12	40	

Table 3 Correlation between disease and exfoliated epithelial membranes ($n = 70$)

Item	*Category*	*Exfoliated epithelial membrane* *No*	*Yes*	*Total*	*p Value*
Cerebral haemorrhage	Yes	9	5	14	0.200
	No	45	11	56	
Dementia	Yes	24	14	38	0.003
	No	30	2	32	
Heart failure	Yes	16	5	21	0.901
	No	38	11	49	
High blood pressure	Yes	24	6	30	0.622
	No	30	10	40	
Ischaemic heart disease	Yes	12	2	14	0.497
	No	42	14	56	
Parkinson's disease	Yes	3	3	6	0.128
	No	51	13	64	
Respiratory disease	Yes	4	2	6	0.614
	No	50	14	64	
Gastrointestinal disease	Yes	14	10	24	0.007
	No	40	6	46	
Kidney disease	Yes	4	2	6	0.614
	No	50	14	64	
Diabetes	Yes	8	2	10	1.000
	No	46	14	60	
Hyperlipidaemia	Yes	4	0	4	0.567
	No	50	16	66	
Bone and joint disease	Yes	19	0	19	0.004
	No	35	16	51	
Skin disease	Yes	6	8	14	0.001
	No	48	8	56	
Epilepsy	Yes	9	5	14	0.200
	No	45	11	56	
Malignant tumour	Yes	8	0	8	0.184
	No	46	16	62	

dryness of the tongue dorsum and open mouth were observed to correlate with the formation of a peeling epithelial membrane. The percentage of misclassification was 11.5%.

The factor with the highest correlation to the formation of membranous substances was the method of nutrient ingestion with 40% of the tube-fed patients in node 1 having peeling epithelial membranes. On the other hand, peeling epithelial membranes were not observed among any tube-fed patients in node 2. The factor with the second highest correlation to membrane formation was dryness of the tongue dorsum. Among tube-fed patients, no peeling epithelial membranes were observed in patients in node 3 who had dampness of the tongue dorsum, while 82.4% of the patients in node 4 with dryness of the tongue dorsum were positive for peeling epithelial membranes. The third highly correlated factor was open mouth, in which the patients' mouths are open at all times. Among the patients in node 5 who had open mouth, two were found to have peeling epithelial membranes. No peeling of the epithelial membrane was observed in those patients without an open mouth.

Adjusted odds ratios

Of the three factors obtained through the decision tree analysis, oral ingestion/intubation, which was most highly correlated with formation of the membranes, could not be used for logistic regression analysis because no patients with oral ingestion showed formation of peeling epithelial membranes. Therefore, the oral ingestion/intubation variable was deleted from the model, and logistic regression was carried out using the factors dryness of the tongue dorsum (wettability

Table 4 Correlation between common drugs and exfoliated epithelial membranes (n = 70)

Item	*Category*	*Exfoliated epithelial membrane* *No*	*Yes*	*Total*	*p Value*
Ca antagonist	Yes	11	0	11	0.058
	No	43	16	59	
ACE inhibitor	Yes	6	2	8	1.000
	No	48	14	62	
β Blocker	Yes	1	0	1	1.000
	No	53	16	69	
α Blocker	Yes	7	1	8	0.672
	No	47	15	62	
Administration of antihypertensives	Yes	17	3	20	0.529
	No	37	13	50	
Cyclic improving agent	Yes	2	1	3	0.547
	No	52	15	67	
Peptic ulcer agent	Yes	16	6	22	0.555
	No	38	10	48	
Antiparkinson agent	Yes	3	0	3	1.000
	No	51	16	67	
Antipodagric	Yes	2	0	2	1.000
	No	52	16	68	
Antiepileptic agent	Yes	12	3	15	1.000
	No	42	13	55	
Hypoglycaemic agent	Yes	2	0	2	1.000
	No	52	16	68	
Electrolytic preparation	Yes	1	1	2	0.407
	No	53	15	68	
Bowel function regulator	Yes	1	1	2	0.407
	No	53	15	68	
Purgative	Yes	10	4	14	0.723
	No	44	12	56	
Lactobacillus preparation	Yes	10	11	21	<0.001
	No	44	5	49	
Expectorant	Yes	7	8	15	0.002
	No	47	8	55	
Diuretic	Yes	12	4	16	1.000
	No	42	12	54	
Antibacterial agent	Yes	2	7	9	<0.001
	No	52	9	61	
Nitric acid medicine	Yes	7	2	9	1.000
	No	47	14	61	
Antiplatelet agent	Yes	12	1	13	0.272
	No	42	15	57	
Anticoagulant	Yes	3	0	3	1.000
	No	51	16	67	
Lactose	Yes	1	3	4	0.035
	No	53	13	66	
Vitamin compound	Yes	11	1	12	0.272
	No	43	15	58	
Iron preparation	Yes	4	0	4	0.567
	No	50	16	66	
Chinese herbal medicine	Yes	2	1	3	0.547
	No	52	15	67	
Luteal hormone	Yes	1	0	1	1.000
	No	53	16	69	