We examined the amlodipine dissolution from orally disintegrating tablets (ODTs) in vivo in the human oral cavity. Additionally, 5 different in vitro short dissolution test methods (Tricorptester, magnetic stirrer, rotating injection syringe, paddle apparatus, shaking) were used to evaluate dissolution and the results were compared to those obtained with the human volunteers. Various amlodipine ODTs with different levels of physical masking effectiveness were manufactured using the RACTAB® technique. Quantitative findings showed that amlodipine dissolution from ODT was dependent on time in the oral cavity and the amount of coating applied for physical masking. We also found that dissolution in the oral cavity was best correlated to that in in vitro short dissolution tests with a time period of 30 s. For more detailed evaluations, mean prediction error, mean absolute error, and root mean square error values were calculated, each of which was lowest with the Tricorptester method among all of the investigated test methods. Our results indicate that mimicking the inside of the human oral cavity is accurate with a testing time of 30 s, while the Tricorptester method was the most preferable of all in vitro tests investigated in this study.

Key words: amlodipine; orally disintegrating tablet; dissolution; oral cavity; bitterness masking; in vitro short dissolution test

Orally disintegrating tablets (ODTs) were actively developed in the late 1980 and several related products have since been introduced,1) with benefits noted for patients with dysphagia as well as improvements in drug regimen adherence.2) An important characteristic of ODTs is a rapid disintegration time in the oral cavity. Indeed, according to guidelines issued by the United States Food and Drug Administration, the required disintegration time for these tablets is 30 s or less.3) Since their introduction, pharmaceutical companies have been developing high-functional ODTs with a rapid disintegration time by making use of a variety of formulation techniques4) and have conducted in vitro tests to evaluate the time required for disintegration.5)

When an ODT includes a bitter tasting active pharmaceutical ingredient (API), a part of that ingredient is dissolved in the oral cavity, which can cause discomfort for the patient and affect drug regimen adherence.2) Thus, it is necessary to mask the bitter taste as much as possible and a water insoluble polymer coating, termed physical masking, is often used.5,6) The purpose of bitterness masking is to reduce API dissolution to a low level so that the patient is insensitive to the bitter taste in the oral cavity when taking an ODT. Therefore, determination of the amount of API dissolution in the oral cavity is an important issue, though no such findings have been reported thus far.

A variety of in vitro methods for mimicking the human oral cavity have been researched and developed, such as the modified paddle apparatus,7–9) rotating injection syringe,10–12) injection syringe combined with delivery pump,13) magnetic stirrer,14) touch mixer,15) mini-column,16) and glass tube17) methods. However, none of those have been verified to produce results that are comparable to API dissolution in the oral cavity.

On the other hand, it is considered that a human gustatory sensation test is more effective to evaluate the bitterness of ODTs, though that is generally an undesirable examination. Furthermore, if the examined ODT includes an API for which safety and potential toxicity are unknown, human testing must be avoided on ethical grounds. Therefore, establishment of an effective in vitro evaluation method that mimics the human oral cavity is considered to be important.

The aim of the present study was to clarify API dissolution in the oral cavity. We selected amlodipine besylate as a model API because of its bitterness and manufactured several types of granules that included amlodipine and ethyl cellulose, a water insoluble polymer, as the physical masking agent. Using the RACTAB® technique, we manufactured various amlodipine ODTs composed of granules that were different in regard to physical masking effectiveness.18,19) They were given to healthy volunteers and we evaluated API dissolution in the oral cavity, including the relationship between in vivo dissolution and the amount of coating used for physical masking. Additionally, several in vitro short dissolution tests were used to evaluate amlodipine dissolution and the results were compared to those obtained with the human volunteers.
Experimental

Materials Amlodipine besylate was purchased from Daito Co., Ltd. (Toyama, Japan) and used as a bitter drug model. Ethyl cellulose (Dow Chemical Co., Ltd., Tokyo, Japan) and Talc (Hayashi Kasei Co., Ltd., Osaka, Japan) were used as coating agents. ß-Mannitol (Roquette Japan Co., Ltd., Tokyo, Japan), light anhydrous silicic acid (Fuji Siyusya Chemical Co., Ltd., Tokyo, Japan), and magnesium stearate (Taihei Chemical Industrial Co., Ltd., Osaka, Japan) were used as excipients for tablet formulation. Rapid disintegration granules, a key component of the RACT AB® technique for causing rapid ODT disintegration, was kindly provided by Towa Pharmaceutical Co., Ltd. (Osaka, Japan). All raw materials obtained were Japanese pharmacopoeia or pharmaceutical excipient grade chemicals.

Preparation of ODTs Amlodipine besylate and ß-mannitol were granulated and coated with the liquid containing solved ethyl cellulose and dispersed talc in a fluidized-bed granulator (MP-01, Powrex Corp., Hyogo, Japan).

The volume of ethyl cellulose used for the amlodipine coating varied, with 4, 6, and 10% of the whole amlodipine ODT mass used to manufacture C4, C6, and C10 granules, respectively. Granules without ethyl cellulose or talc as a coating agent were also prepared by mixing amlodipine besylate and ß-mannitol, and denoted as C0 granules.

Granules including amlodipine besylate (C0, C4, C6, C10), rapid disintegration granules, ß-mannitol, light anhydrous silicic acid, and magnesium stearate were mixed to manufacture 4 different kinds of granules for tableting. Using a rotary tableting machine (VELA5, Kikusui Seisakusho Ltd., Kyoto, Japan), amlodipine ODTs including those composed of C0, C4, C6, and C10 granules were manufactured to produce C0-ODT, C4-ODT, C6-ODT, and C10-ODT, respectively. The weight and diameter of each ODT was 205 mg and 8 mm, respectively. The characteristics of the amlodipine ODTs used in this study are shown in Table 1.

Amlodipine Dissolution from ODTs in Human Oral Cavity This study was conducted in accordance with the Declaration of Helsinki, as noted by the World Medical Association, and Ethical Guidelines for Clinical Research from the Ministry of Health, Labour and Welfare in Japan. The study protocol was approved by the Ethics Committee of Hamamatsu University School of Medicine, Japan. The study was registered in the UMIN Clinical Trials Registry (UMIN000030831). Ten healthy volunteers [9 men, 1 woman; mean age±standard deviation (S.D.) 22.7±0.8 years] participated as subjects after providing written informed consent.

This study was conducted as a randomized crossover single-blinded trial and the test procedures are shown in Fig. 1. Each subject placed an amlodipine ODT in their mouth. After 15, 30, or 60 s they spat out the disintegrated ODT along with saliva into an injection syringe (SS-20ESZ, Terumo Corp., Tokyo, Japan), then those contents were immediately passed through a disc filter (25CS045AN, Advantec Toyo Kaisha Ltd., Tokyo, Japan) into a glass tube, with any remnants in the oral cavity soon removed with gauze (No. 5050, Yamatokojo Co., Ltd., Osaka, Japan). To return their oral cavity to the original state and prepare for the next ODT, the subjects rinsed their mouth with 150 mL of mineral water twice, with the next test conducted 20 min after the previous. The amlodipine concentration in saliva was determined using HPLC. In addition, the injection syringe, gauze, and glass tube used were weighed before and after each test to calculate the total saliva amount for each test (1 mg=1 mL).

Table 1. Characteristics of Amlodipine ODTs

<table>
<thead>
<tr>
<th></th>
<th>C0-ODT</th>
<th>C4-ODT</th>
<th>C6-ODT</th>
<th>C10-ODT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hardness (N)</td>
<td>57.3±1.2</td>
<td>72.0±2.2</td>
<td>72.0±0.8</td>
<td>70.3±2.1</td>
</tr>
<tr>
<td>Disintegration time (s)</td>
<td>12.9±0.3</td>
<td>11.5±0.5</td>
<td>11.0±0.4</td>
<td>12.3±0.4</td>
</tr>
<tr>
<td>Dissolution ratio (%)</td>
<td>5 min</td>
<td>74.8±4.2</td>
<td>67.7±1.5</td>
<td>62.5±2.7</td>
</tr>
<tr>
<td></td>
<td>15 min</td>
<td>86.5±3.2</td>
<td>77.6±1.3</td>
<td>76.0±0.6</td>
</tr>
<tr>
<td></td>
<td>30 min</td>
<td>90.5±1.7</td>
<td>80.0±0.9</td>
<td>78.4±0.7</td>
</tr>
</tbody>
</table>

Values are shown as the mean±S.D. (n=3). Hardness was measured with a hardness meter (PC-30, Okada Seiko Co., Ltd., Tokyo, Japan) and disintegration time with a Tricorp-tester (Okada Seiko Co., Ltd.). The dissolution ratio of amlodipine from the ODTs was measured using a paddle apparatus (50 rpm, 900 mL water).

Fig. 1. Method and Time Course of in Vivo Dissolution Test
In Vitro Short Dissolution Test  For the in vitro tests, we used 15-, 30-, and 60-s time periods, the same as in the in vivo test method shown above. Obtained test liquid samples were filtered using a disc filter (GLCTD-MCE2545, Shimadzu Glc Ltd., Tokyo, Japan). Amlodipine concentration in each test liquid sample was determined using HPLC. Each test was conducted in triplicate.

Tricorptester Method  For this method, we used a Tricorptester device (Okada Seiko Co., Ltd., Tokyo, Japan) as the ODT disintegration tester and also considered it for use as a dissolution tester. This device has 2 metal mesh screens to hold an ODT on the upper and lower sides. When conducting a test, artificial saliva (NaCl, 1.44 g/L; KCl, 1.47 g/L; and Tween 80, 0.3%) at 37°C was dripped from a height of 80 mm at a rate of 3.0 mL/min onto the ODT. The default setting for the device stops the artificial saliva from dripping when the ODT shows complete disintegration and the 2 mesh screens touch each other. However, for the present study, the artificial saliva dripping was continued until the end of the testing time period even if the ODT had completely disintegrated.

Magnetic Stirrer Method  This method was conducted with use of a magnetic stirrer (RS-1DN, AS ONE Corp., Osaka, Japan). The dissolution medium was 10 mL of purified water at 37°C in a 30-mL beaker (Hario Science Co., Ltd., Tokyo, Japan). That was placed on the magnetic stirrer, then the device rotator was placed inside the beaker and rotated at 200 rpm.

Rotating Injection Syringe Method  We used a method previously reported by Shirai et al. The dissolution medium was 5 mL of purified water at 37°C in a 10-mL injection syringe (SS-10SZ, Terumo Corp.). That was rotated once every 3 s.

Paddle Apparatus Method  For this method, we used a dissolution tester (PJ-6S, Miyamoto Riken Industry Co., Ltd., Osaka, Japan). The dissolution medium was 900 mL of purified water at 37°C and the paddle rotation speed was 50 rpm.

Shaking Method  This test was performed with a shaking machine (PERSONAL-11, Taitec Corp., Saitama, Japan). The temperature of the water bath was set at 37°C, then 10 mL of purified water used as the dissolution medium was poured into a 30-mL conical flask (Hario Science Co., Ltd.) and that was placed into the bath, with the shaking speed set at 100 rpm.

HPLC Condition  The concentration of amlodipine besylate was determined using an HPLC system (Prominence UFLC, Shimadzu Corporation, Kyoto, Japan) comprised of a pump (LC-20AD, Shimadzu Corporation), online degassing unit (DGU-20A3, Shimadzu Corporation), column oven (CTO-20AC, Shimadzu Corporation), autosampler (SIL-20AC, Shimadzu Corporation), and photodiode array detector (SPD-M20A, Shimadzu Corporation), which were integrated by use of a system controller (CBM-20A, Shimadzu Corporation). HPLC was performed using an analytical column (Atlantis

Fig. 2. Dissolution Ratio of Amlodipine in Oral Cavity
(a) C0-ODT, (b) C4-ODT, (c) C6-ODT, (d) C10-ODT. Each point represents the dissolution ratio of amlodipine in the oral cavity of the 10 healthy volunteers (9 men, 1 woman; mean age±S.D.=22.7±0.8 years). Horizontal line indicates the mean value for each test time period.
column, 4.6 × 150 mm, Waters Corporation, Tokyo, Japan) with a mobile phase (0.05 mM triethylamine (pH 3.0): acetonitrile=3:2) delivered at a flow rate of 1 mL/min. The column temperature was maintained at 40°C and detection was based on UV absorbance at 237 nm.

**Evaluation of Consistency of Dissolution in the Oral Cavity** We evaluated the consistency of dissolution of amiodipine between the human subjects and in the 5 different *in vitro* short dissolution test methods to determine which of those methods best mimicked dissolution in the oral cavity. Mean prediction error (MPE), mean absolute error (MAE), and root mean square error (RMSE) values were calculated using the residual of the dissolution ratio in the oral cavity ($D_{vivo}$) and that in the *in vitro* short dissolution test ($D_{vitro}$), with $D_{vivo}$ was used as a reference, with the results expressed as a percentage (%MPE, %MAE, %RMSE, respectively).

\[
%\text{MPE} = \frac{1}{n} \sum_{i=1}^{n} \left( \frac{1}{D_{vivo}} (D_{vivo} - D_{vivo}) \times 100 \right)
\]

\[
%\text{MAE} = \frac{1}{n} \sum_{i=1}^{n} \left( \frac{1}{D_{vivo}} (D_{vivo} - D_{vivo}) \times 100 \right)
\]

\[
%\text{RMSE} = \sqrt{\frac{1}{n} \sum_{i=1}^{n} \left( \frac{1}{D_{vivo}} (D_{vivo} - D_{vivo}) \times 100 \right)^2}
\]

**Statistical Analysis** All values are shown as the mean±S.D. Statistical analysis was performed using the Graphpad Prism v. 5.02 software package (Graphpad Software, San Diego, CA, U.S.A.).

**Results**

**Amlodipine Dissolution in Oral Cavity of Healthy Subjects** The dissolution ratios of 4 different amiodipine ODT formulations (C0-ODT, C4-ODT, C6-ODT, C10-ODT) in the oral cavity of the 10 subjects are shown in Fig. 2. The mean dissolution ratio tended to increase over time. As the coating amount of ethyl cellulose increased, the mean dissolution ratio of amiodipine for each testing time was decreased. Additionally, we found no differences in mean saliva amount among the different amiodipine ODT formulations (Table 2).

**Amlodipine Dissolution in *in Vitro* Short Dissolution Tests** Five different methods were used to conduct *in vitro* testing and the results are shown in Fig. 3. In each of the tests, the mean dissolution ratio of amiodipine tended to increase over time, though the value obtained with the shaking method was much higher than that obtained with the other methods.

**Table 2. Saliva Amount in Subjects for Each Testing Time**

<table>
<thead>
<tr>
<th>Testing time (s)</th>
<th>C0-ODT</th>
<th>C4-ODT</th>
<th>C6-ODT</th>
<th>C10-ODT</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>1.55±0.64</td>
<td>1.46±0.39</td>
<td>1.43±0.43</td>
<td>1.44±0.47</td>
</tr>
<tr>
<td>30</td>
<td>1.71±0.63</td>
<td>1.75±0.52</td>
<td>1.75±0.62</td>
<td>1.78±0.64</td>
</tr>
<tr>
<td>60</td>
<td>2.31±0.74</td>
<td>2.37±0.88</td>
<td>2.17±0.71</td>
<td>2.12±0.71</td>
</tr>
<tr>
<td>Overall</td>
<td>1.51±0.49</td>
<td>1.75±0.61</td>
<td>2.24±0.77</td>
<td></td>
</tr>
</tbody>
</table>

Saliva amounts are presented as mL and shown as the mean±S.D. (n=10).

Fig. 3. Dissolution Ratio of Amlodipine in *in Vitro* Short Dissolution Tests

(a) Tricorptester, (b) magnetic stirrer, (c) rotating injection syringe, (d) paddle apparatus, (e) shaking. ● C0-ODT, ■ C4-ODT, ▲ C6-ODT, ▼ C10-ODT. The dissolution ratio for each time period is shown as the mean±S.D. (n=3).
As the ethyl cellulose coating amount increased, the mean dissolution ratio of amlodipine decreased with all testing times, except with the paddle apparatus method.

**Difference in Dissolution between Human Subjects and in Vitro Short Dissolution Testing** The dissolution ratio of amlodipine in the human oral cavity and different in vitro short dissolution tests are shown in Fig. 4. In that figure, a line with a slope of 1 was drawn from the original position 0 to clearly show the different dissolution ratios between the values being compared in each graph. We found that the amount of dissolution after 15 s in the human subjects tended to be greater than that in the in vitro short dissolution tests, whereas that amount in the in vitro tests tended to be greater than in the human subjects after 60 s. After 30 s, there was no evident

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![Fig. 4. Dissolution Ratio of Amlodipine in Oral Cavity (in Vivo) Compared to That in in Vitro Short Dissolution Tests](image)

(a) Tricorptester, (b) magnetic stirrer, (c) rotating injection syringe, (d) paddle apparatus, (e) shaking.  15 s, ▲ 30 s, ■ 60 s.

**Table 3. Consistency of Different Methods with Dissolution in Oral Cavity after 15, 30, and 60 s**

<table>
<thead>
<tr>
<th></th>
<th>r</th>
<th>%MPE</th>
<th>%MAE</th>
<th>%RMSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 s</td>
<td>Tricorptester</td>
<td>0.929</td>
<td>−70.8 (−83.8–−57.7)</td>
<td>70.8 (57.8–83.8)</td>
</tr>
<tr>
<td></td>
<td>Magnetic stirrer</td>
<td>0.918</td>
<td>−39.6 (−69.9–−9.36)</td>
<td>45.3 (24.1–66.5)</td>
</tr>
<tr>
<td></td>
<td>Rotating injection syringe</td>
<td>0.957</td>
<td>−47.2 (−66.5–−27.9)</td>
<td>47.2 (27.9–66.5)</td>
</tr>
<tr>
<td></td>
<td>Paddle apparatus</td>
<td>0.156</td>
<td>−94.5 (−100.1–−89.1)</td>
<td>94.6 (89.1–100.1)</td>
</tr>
<tr>
<td></td>
<td>Shaking</td>
<td>0.955</td>
<td>−5.1 (−39.1–28.8)</td>
<td>30.2 (12.9–47.6)</td>
</tr>
<tr>
<td>30 s</td>
<td>Tricorptester</td>
<td>0.941</td>
<td>−0.8 (−21.7–20.1)</td>
<td>17.4 (5.4–29.5)</td>
</tr>
<tr>
<td></td>
<td>Magnetic stirrer</td>
<td>0.875</td>
<td>−21.7 (−57.7–14.2)</td>
<td>40.9 (29.3–52.6)</td>
</tr>
<tr>
<td></td>
<td>Rotating injection syringe</td>
<td>0.932</td>
<td>−12.5 (−52.5–27.5)</td>
<td>38.1 (19.3–56.9)</td>
</tr>
<tr>
<td></td>
<td>Paddle apparatus</td>
<td>0.949</td>
<td>−34.6 (−60.1–−9.2)</td>
<td>34.6 (9.2–60.1)</td>
</tr>
<tr>
<td></td>
<td>Shaking</td>
<td>0.977</td>
<td>40.5 (1.7–79.3)</td>
<td>47.8 (17.9–77.6)</td>
</tr>
<tr>
<td>60 s</td>
<td>Tricorptester</td>
<td>0.859</td>
<td>39.2 (−98.8–88.2)</td>
<td>48.0 (7.2–88.8)</td>
</tr>
<tr>
<td></td>
<td>Magnetic stirrer</td>
<td>0.908</td>
<td>6.4 (−31.0–43.8)</td>
<td>27.2 (0.3–54.2)</td>
</tr>
<tr>
<td></td>
<td>Rotating injection syringe</td>
<td>0.922</td>
<td>20.8 (−21.6–62.8)</td>
<td>38.8 (11.7–65.9)</td>
</tr>
<tr>
<td></td>
<td>Paddle apparatus</td>
<td>0.788</td>
<td>27.2 (−10.2–64.5)</td>
<td>38.8 (13.1–64.5)</td>
</tr>
<tr>
<td></td>
<td>Shaking</td>
<td>0.983</td>
<td>63.3 (31.0–95.6)</td>
<td>63.3 (31.0–95.6)</td>
</tr>
</tbody>
</table>

r, correlation coefficient; %MPE, percent mean prediction error; %MAE, percent mean absolute error; %RMSE, percent root mean square error. Values are shown as the mean (95% coefficient interval, n=4).
difference between the in vivo and in vitro. The values for the correlation coefficient at 30 s were greater than 0.875. For more detailed evaluations, %MPE, %MAE, and %RMSE values were calculated. These values of 15 and 60 s were higher than those of 30 s, except for the shaking method at 15 s and the magnetic stirrer method at 60 s. All were lowest with the Tricorptester method after 30 s (Table 3).

Discussion

The aim of this study was to clarify the amount of amlodipine dissolution from ODT in the oral cavity of human subjects. We selected amlodipine besylate as the model API, then prepared ODT including non-coated amlodipine besylate (C0-ODT) and ODTs with granules including coated amlodipine (C4-ODT, C6-ODT, C10-ODT). Amlodipine dissolution from these ODTs was examined and our results showed that the amount increased in accordance with time after taking the ODT. Additionally, increased levels of ethyl Cellulose coating amount were shown to suppress dissolution in the oral cavity, indicating the effectiveness of bitterness masking.

In a previous study that conducted human gustatory sensation tests using amlodipine ODTs, differences between the presence and absence of ethyl cellulose coating used for bitterness masking were noted. In that study, ODTs were manufactured with the same ingredients and method as in the present investigation, then a visual analog scale (VAS) was utilized for evaluation, with a larger VAS value considered to indicate better palatability. Their results confirmed that ODTs with coated amlodipine besylate had a higher level of palatability as compared to non-coated ODTs. On the other hand, our findings showed that amlodipine dissolution in the oral cavity was decreased as the amount of ethyl cellulose coating increased. Both studies suggested that a decrease in dissolution in the oral cavity was correlated with improvement in palatability. The present results are the first to quantitatively show that to coat API as bitterness masking improves palatability.

We also investigated in vitro short dissolution test methods for mimicking amlodipine dissolution from ODT in the oral cavity. A variety of studies have used magnetic stirrer, rotating injection syringe, paddle apparatus, and shaking methods, while the Tricorptester employed in the present examination is a relatively new device. We found that all except for the paddle apparatus method resulted in a decreased dissolution ratio in association with an increase in coating amount, which agreed with the results obtained in the human subjects. Thus, we consider that these in vitro methods, except for the paddle apparatus, are suitable for short dissolution testing for confirming effectiveness in quality tests of pharmaceutical preparations.

When we focused attention on the testing period, the dissolution ratio was found to increase with time. Interestingly, the amount of dissolution in the oral cavity after 15 s in the human subjects tended to be higher as compared to that seen with the in vitro methods, whereas dissolution in the in vitro test results tended to be lower than that in the in vivo examination after 60 s. Because of the short testing times used in our study, we considered that ODT disintegration was insufficient and could lead to a delay in in vitro amlodipine dissolution. Conversely, with an extended testing time, we speculated that unnecessary pressure would be added to the ODT during in vitro short dissolution testing. In the human subjects, the ODT was subjected to pressure in the oral cavity until complete disintegration, then that pressure was discontinued until the end of the test period. On the other hand, with the in vitro short dissolution tests, pressure continued to be applied until the end of the designated time period even if the ODT had completely disintegrated.

Generally, most ODTs are formulated to disintegrate within 30 s. In this study, the difference regarding dissolution ratio between the in vivo and in vitro tests seemed to be minimized with the 30-s period, indicating the reasonableness of that amount of time. For statistical analysis, we calculated %MPE, %MAE, and %RMSE values. Those obtained with the Tricorptester method were lowest and this method was considered to more precisely mimic dissolution in the oral cavity of all the testing methods examined in this study. Therefore, we concluded that the dissolution ratio in in vitro short dissolution tests was comparable to that in the oral cavity with a testing time of 30 s. Furthermore, of all in vitro tests examined in the present study, the Tricorptester method was the most precise for mimicking the inside of the human oral cavity.

Conclusion

The present results are the first to show that drug dissolution from ODT is dependent on time in the oral cavity and coating amount. We also found that dissolution in the oral cavity was best correlated to that in in vitro short dissolution tests when a time of 30 s was used. In addition, the %MPE, %MAE, and %RMSE values obtained with the Tricorptester method were lowest among all of the investigated test methods. Our results suggest the possibility to expand the methods utilized to examine dissolution in the oral cavity.

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Conflict of Interest T.K. is an employee of Towa Pharmaceutical Co., Ltd. (Osaka, Japan). S.U. and N.N. received research Grants from Kissei Pharmaceutical Co., Ltd., Takeda Consumer Healthcare Co., Ltd. (Tokyo, Japan), and Otsuka Pharmaceutical Co., Ltd. (Tokyo, Japan). N.N. serves as a consultant to Kissei Pharmaceutical Co., Ltd., Otsuka Pharmaceutical Co., Ltd., and Shiseido Japan Co., Ltd. (Tokyo, Japan). None of the authors have conflicts of interest to declare in regard to this study.

References