

Short-Term Effectiveness of a Swallowing Exercise for the Elderly Using Day-Care Services

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Abstract

Purpose: The research purpose was to investigate the short-term effectiveness of exercise intervention to improve pharyngeal functions in the elderly using day-care services.

Methods: Subjects were totally 84 elderly people. The participants were analyzed at 2 points: baseline and 2 weeks after the starting date of the study. This exercise comprised pronunciation practice, a pushing exercise with crying out in a loud voice, and a falsetto exercise. A questionnaire of ingestion and deglutition functions was also answered at 2 points. The collected data were statistically analyzed using SPSS J16.0. The intervention duration of this study was from February 2 to March 4, 2008.

Results: The average participant age was 82.4 ± 8.6 years. Comparing the evaluative items of the swallowing functions at 2 points, significant differences were observed in the movement of the upper and lower sides of the tongue, optional cough, consecutive pronunciation [ta], hoarseness, and the RSST test. The total frequency of participation in this programmed exercise was significantly related to lip and tongue movement, pronunciation [ka], and utterance time. A positive change of feeling was observed in the score of the 2 items. **Conclusion:** The results of this study showed very useful hypothetical information about intervention to arouse pharyngeal functions for the elderly using day care services.

Keywords: Effectiveness; Swallowing; Exercise; The elderly; Day care service

Introduction

The 5 swallowing stages consist of perceptive, pre-oral, oral, pharyngeal and esophageal, and this process continuously works to transport the bolus of food and drink. Dysphagia is usually caused by some diseases such as stroke, neurological diseases, cancer and so on. However, the elderly has some physiological hypofunctions on normal swallowing pattern, as neuromuscular reserve reduces in aging [1].

In the elderly muscle volume of the tongue gradually reduces [2] and the tongue force of transferring bolus from the oral stage to the pharyngeal stage decreases [3]. Furthermore, the bolus transportation time in the pharyngeal stage is slightly longer than normal [4-9], as the pharyngeal wall contraction weakens, small muscle volume around the pharynx decreases [8,9] and the timing of triggering reflex gets slow [1,10-13]. Additionally, the frequency of the residue in the elderly increases approximately 2-3% compared with that in younger people [13], because the position of the larynx in some elderly people shifts downward [1,14], the cavity of pharynx spreads [1,5,15], the opening of the upper esophagus sphincter (UES) decreases during the bolus passes [1,14] and the timing of UES opening delayed slightly [4,13]. The unusual residue in the pharynx is sometimes aspirated into lung, if the closure of vocal cords also worsens with age [16] and the cough reflex weakens.

Previous studies reported that healthy elderly with dysphagia was around 15% [17,18]. In this research the subjects were frail elderly using day care service of one of national public welfare insurance services to support their physical functional decline. All users of these services need to be judged an appropriate level of their physical and mental conditions before start through official staff and committee. Generally, the permitted users gather to institution to get service in daytime, if they require. In this day care service oral and respiratory exercise is already conducted. However, the menus of improving pharyngeal function to prevent aspiration have never included. Then, we expected intentional intervention to strengthen pharyngeal muscles using existing exercise could lead positive outcomes in the elderly.

There are many specific training menus for actual dysphagic clients in acute clinical settings to remain and improve the tongue muscles, larynx movement, vocal cord adduction, respiration, and so on [19]. These menus aim improvement of bolus transportation by tongue training [19-20], prevention of aspiration by strengthening a closure of the glottis using pushing exercises [19,21-23], improvement of pharynx contraction and upward larynx movement using falsetto voice training (speaking in the highest possible voice) [19], strengthening of suprahyoid muscles and increasing larynx upward movement by Shaker exercises [24-31], improvement of coordination between swallowing and breathing based on stretching of the intercostal muscles, and improvement of expiration capability at the time of aspiration using the Sylvester method [31]. For the frail elderly, a motionless or lower motion naturally reduces entire muscle volume and stimulating swallow related muscles is important for first preventive rehabilitation as same as keeping walk ability. Therefore, we investigators selected some additional appropriate exercises from the existing rehabilitation menus for the frail elderly at a high risk of swallowing disorder to promote pharyngeal functions, and tried to intervene in daycare setting.

The purpose of this research was to estimate the short-term effectiveness of exercise intervention to improve the swallowing function among the elderly using daycare service.

Methods

Eighty-four elderly people (80.0%) of all 105 registered users in a single institution joined in this research. Exclusive criterion was the user

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with severe communication disorder or paralysis. A number was 2. The participants were analyzed at 2 points: baseline (before) and 2 weeks after the starting date of the study. Before starting the process, the participants were informed about the purpose of this study, and they provided their consent. This study was approved by an ethical board of study institution. The content of intervention based on fundamental knowledge in the literature and books on rehabilitation for dysphagic patients was developed by researchers and speech-language pathologist (SLP).

Every time the subjects participated in day care service, they were asked to participate in intervention exercise. A questionnaire of this study including ingestion and deglutition functions was answered by participations at baseline and after intervention. The questionnaire asked about characteristics, the long-term care insurance level [32], Barthel index (BI) [33,34], Karasawa dementia scale [35], ingestion and deglutition functions such as repetitive saliva swallowing test (RSST) [36,37], cough test (CT) [38], the related contents of the medical observations of dysphagia, part of the lower rank items of disease-specific questionnaire measuring dysphagia patients' experiences of health outcomes (the SWAL-QOL) [39,40], and others. Long-term care insurance is provided under Japanese law, and the level of the insurance depends on the actual care necessity for elderly people [34]. The answers to the long-term care insurance level, dementia level, and ingestion and deglutition functions were classified as the Likert scale, while the BI score ranged from 0 to 100.

The intervention program, which was added the new contents related improvement of pharyngeal functions, was conducted by the staffs in the institution. Basic usual exercise was comprised pronunciation practice of [pa][ta][ka][ra][patakara].

Each pronunciation was repeated 3 times at a time per one service day. These pronunciations affect the relaxation or strength of the lips, the front and middle of the tongue, the base of the tongue, the cheeks, and the coordination of those organs [1]. Additionally, a pushing exercise with crying out in a loud voice [ei] to strengthen the closure of vocal cords was added into a new intervention program and carried out 10 times. Furthermore, a falsetto exercise, which was compromised the contents of heightening a scale of pronunciation [i:] and keeping it 5 seconds, was also added and carried out 3 times. These 2 exercises strengthen the muscles surrounding the pharynx and the upward movement of the larynx and have never performed in day care service group. When performing any exercises, it is an important point that adding the defined load to the related muscles under safe. Therefore, the researcher requested that the staffs would instruct forceful and steady exercises with suggestions and check the user's performance. If the users do not perform enough, the staffs guided the user good exercise as possible as they can. Before starting this study, the staffs were trained for demonstration of the intervention exercise to the users according to intervention contents by a dysphagia expert researcher, and they understood how to perform the programed exercise. After those, the collected data were statistically analyzed by SPSS 16.0 Japanese version. The intervention duration of this study was from February 2 to March 4, 2008.

Results

The data of 73 (86.9%) users, excluding the users who dropped out or without some data, were analyzed. The reasons of dropout were absence due to a hospital visit and/or disease from survey day, reluctance to join, and others. The average participant age was 82.4 ± 8.6 years, and the number of female patients was 52 (71.2%). Furthermore, a number of subjects whose ages 80–84 years was 21 (28.8%), while a number of subjects >85 years of age was 21 (29.0%).

Regarding the long-term care insurance level, 26 (35.6%) patients required support level 1 or 2, and the majority of subjects were in these levels. Moreover, a number of subjects who consulted doctors were 70 (95.9%), and the rate of circulatory disease according to the International Classification of Diseases (ICD-10) criteria among all groups was the highest. A number of subjects >90 of BI score was 59 (80.8%). Regarding dementia level, normal or slight dementia was the most common. The highest frequency ratio of the elderly using day care services was once a week at 53.6% (Table 1).

Comparing the evaluative items of the swallowing functions before and after intervention, significant differences were observed in lip projection, optional cough, consecutive pronunciation [ta], hoarseness

	Items	n	%
Gender	Male	21	28.8
	Female	52	71.2
Age	-74	8	10.9
	75-79	16	21.9
	80-84	21	28.8
	85-89	19	26.3
	90-	2	2.7
	(Mean±SD)		82.4 ± 8.6
Adopting Level of long-term care insurance			
	people requiring support 1,2	26	35.6
	people requiring long-term care, level 1	20	27.4
	people requiring long-term care, level 2	12	16.4
	people requiring long-term care, level 3	8	11.0
	people requiring long-term care, level 4	4	5.5
	people requiring long-term care, level 5	3	4.1
Consult a doctor			
	Existence	70	95.9
	Not existence	3	4.1
Main diagnosis (ICD-10)			
	Diseases of the circulatory system	48	65.8
	Diseases of the nervous system	9	12.3
	Diseases of the musculoskeletal system and connective tissue	8	11.0
	Diseases of the digestive system	4	5.5
	Endocrine, nutritional and metabolic diseases	3	4.1
	No data	1	1.4
Ability of daily living (Barthel Index)			
	-55	4	5.5
	60-65	4	5.5
	70-75	3	4.1
	80-85	3	4.1
	90-95	10	13.7
	100	49	67.1
Ability of feeding by one-self			
	Independent	70	95.9
	Needs a part of assistant	3	4.1
Dementia level Φ			
	Normal	55	75.3
	Slight dementia	9	12.3

Meddle dementia and	9	12.3
Foods consistency		
Normal	67	91.8
Soft foods	2	2.7
Cutting foods	2	2.7
Blender	1	1.4
Other	1	1.4
Denture		
Use	55	75.5
Not use	18	24.7
Frequency of using day service		
(Mean ± SD)	1.73 ± 1.02	
1 time/wk	40	54.8
2 times/	19	26.0
3 times/	11	15.1
4 times/	1	1.4
5-times/	2	2.8

Table 1: Characteristics of the subjects (n=73).

(by Wilcoxon's signed-rank test, $p < 0.05$), and the RSST test (by paired t -test and McNemar test, $p < 0.05$) (Table 2).

Furthermore, the total frequency of participation in this programmed exercise was significantly related to lip and tongue movement, pronunciation [ka], and utterance time (by Spearman's rank correlation coefficient, $p < 0.05$).

Regarding patient's subjective feeling before and after intervention, a positive changes were observed in the score of the 2 items "I take longer to eat" and "I choke while eating", but a negative change was observed in the item "I am afraid of choking when I drink liquids" (by Wilcoxon's signed-rank test $p < 0.05$) (Table 3).

Discussion

The results of this study showed useful hypothetical information about dysphagia intervention for the elderly using day care service. We selected a pushing exercise with the simultaneous voice and a falsetto [i:] as new exercise intervention, especially focusing on the pharyngeal stage in the 5 swallowing stages. A pushing exercise and a falsetto [i:] lead to strengthen the closure force of the vocal cords and the movement of the larynx to prevent aspiration [19,41,42].

Items		Possible		Almost possible		Impossible		z value	p value	
Functional Status	Opening/close mouth	before	72	(98.6)	1	(1.4)	0	(0.0)	1.000	0.317
		after	73	(100.0)	0	(0.0)	0	(0.0)		
	Cheeks: puff out	before	68	(93.2)	2	(2.7)	3	(4.1)	-1.732	0.083
		after	70	(95.9)	1	(1.4)	2	(2.7)		
	put back	before	65	(89.0)	3	(4.1)	5	(6.8)	-1.823	0.084
		after	69	(94.5)	1	(1.4)	3	(4.1)		
	Lip: projection	before	63	(86.3)	4	(5.5)	6	(8.2)	-2.332	0.020*
		after	68	(93.2)	4	(5.5)	1	(1.4)		
	drawing	before	67	(91.8)	2	(2.7)	4	(5.5)	-0.324	0.915
		after	68	(93.2)	1	(1.4)	4	(5.5)		
	Tongue: roll	before	64	(87.7)	3	(4.1)	6	(8.2)	-1.964	0.050
		after	71	(97.3)	0	(0.0)	2	(2.7)		
	forward/backward	before	69	(94.5)	4	(5.5)	0	(0.0)	-1.518	0.180
		after	72	(98.6)	1	(1.4)	0	(0.0)		
	left/right	before	72	(98.6)	0	(0.0)	1	(1.4)	0.000	1.000
		after	72	(98.6)	0	(0.0)	1	(1.4)		
	upward/downward	before	64	(87.7)	6	(8.2)	3	(4.1)	-1.814	0.317
		after	64	(87.7)	9	(12.3)	0	(0.0)		
	Optional cough	before	68	(93.2)	3	(4.1)	2	(2.7)	-2.121	0.034*
		after	72	(98.6)	1	(1.4)	0	(0.0)		
	Pronunciation: [pa]	before	72	(98.6)	1	(1.4)	0	(0.0)	1.000	0.317
		after	72	(98.6)	0	(0.0)	1	(1.4)		
	[ta]	before	71	(97.3)	2	(2.7)	0	(0.0)	-1.342	0.317
		after	72	(98.6)	1	(1.4)	0	(0.0)		
[ka]	before	70	(95.9)	2	(2.7)	1	(1.4)	-1.342	0.180	
	after	72	(98.6)	1	(1.4)	0	(0.0)			
[pa] continuous 5 times	before	71	(97.3)	2	(2.7)	0	(0.0)	1.000	0.317	
	after	71	(97.3)	1	(1.4)	1	(1.4)			
[ta] continuous 5 times	before	62	(84.9)	7	(9.6)	4	(5.5)	-2.486	0.013*	
	after	69	(94.5)	6	(8.2)	1	(1.4)			
[ka] continuous 5 times	before	63	(86.3)	6	(8.2)	4	(5.5)	-1.100	0.271	
	after	65	(89.0)	6	(8.2)	2	(2.7)			
[pataka] continuous 3 times	before	66	(90.4)	4	(5.5)	3	(4.1)	0.000	0.107	
	after	69	(94.5)	4	(5.5)	0	(0.0)			
[a:] ¹	before	73	(100.0)	0	(0.0)	0	(0.0)	1.000	0.317	

		after	72	(98.6)	1	(1.4)	0	(0.0)		
	[e:]	before	72	(98.6)	0	(0.0)	1	(1.4)	1.000	0.227
		after	72	(98.6)	1	(1.4)	0	(0.0)		
	[u:]	before	73	(100.0)	0	(0.0)	0	(0.0)	-0.447	0.655
		after	72	(98.6)	1	(1.4)	0	(0.0)		
	[i:]	before	59	(80.8)	7	(9.6)	7	(9.6)	-1.217	0.317
		after	62	(84.9)	7	(9.6)	4	(5.5)		
Level of Communication		before	69	(94.5)	4	(5.5)	0	(0.0)	0.000	1.000
		after	69	(94.5)	4	(5.5)	0	(0.0)		
				Exist		Slight exist		Not exist	z value	p value
Hoarseness		before	19	(26.0)	6	(8.2)	48	(65.8)	-2.220	0.015*
		after	8	(11.1)	1	(1.4)	64	(87.7)		
				Normal		Abnormal			-	p value
RSST ¹	(n/% of abnormal score: under 2/30s)	before	41	(56.2)		32	(43.8)			0.031*
		after	51	(69.9)		22	(30.1)			
MWST ²	(n/% of abnormal score: under 3)	before	68	(93.2)		5	(6.8)			1.000
		after	68	(93.2)		5	(6.8)			
FT ³	(n/% of abnormal score: under 3)	before	62	(84.9)		11	(15.1)			0.227
		after	67	(91.8)		6	(8.2)			
CT ⁴	(n/% of abnormal score: under 4/minute)	before	63	(86.3)		10	(13.7)			0.453
		after	65	(89.0)		7	(9.6)			
						Mean ± SD			t value	p value
Functional Status	Time of continuing pronunciation [a:] (seconds)	before				10.51 ± 4.66			-0.442	0.660
		after				10.71 ± 4.45				
RSST ¹	Frequency (times)	before				3.05 ± 1.95			-2.021	0.047*
		after				3.37 ± 1.80				

*Analyses were carried out by Wilcoxon's signed-rank test, McNamar test, and paired t-test.

§1: Repetitive Saliva Swallowing Test, normal score is 3 times and over per 30 seconds

§2: Modified Water Swallowing Test, normal score is 4 or 5.

§3: Food Test, normal score is 4 or 5.

§4: Cough Test, normal score is 5 times cough during 1 minute. Total number of comparative analysis after intervention is 72.

Table 2: Comparison ingestion and deglutition evaluations between 2 points (n=73, n (%))

To assess symptom improvement after intervention, voice clarity and swallowing function tests were observed [19]. Both hoarseness and the RSST score as outcome measurements were significantly improved after intervention. Since hoarseness causes vocal fold bowing [41], functional dysphonia [42], or unilateral vocal fold paralysis among elderly people [41,42], the abovementioned result showed a recovery of vocal cord physical status or function. Furthermore, the RSST test is consisted of the oral function, the pharyngeal function and the coordination between swallowing and respiration. The result of this test is used to judge whether aspiration has occurred [36,37]. The result of this test revealed that the average frequency of swallowing increased and that 13.7% in the normal range increased. Although the function of using the tongue and pharynx that participated in the rise of the score by the RSST could not be discerned, the results showed a decrease of aspiration risk after intervention.

In the intervention, we instructed the elderly to conduct a pushing exercise using a loud voice requiring rapid inspiration and expiration. A reflexive cough, which rapidly contracts the diaphragm and the intercostals muscles, is also an important ability of a living body to discharge a foreign body from the respiratory tract or pharynx. In this research, the feasible percentage of an optional cough increased significantly, although the score of the CT used to observe the level of cough reflex triggering did not change. In both cases, muscle flexibility is needed to carry out the rapid inspiration and expiration. In these points the results showed functional improvement. Additionally, deep and rapid respiration is also a very important load exercise for the

elderly to maintain the flexibility of the diaphragm and the intercostal muscles and to improve air capacity. However, we thought that it was difficult to explain to the elderly about how to precisely practice these exercises in our clinical experiments. For the elderly, a pushing exercise with the addition of the loud voice is easier to perform muscle contractions in the pharynx and unconscious effective respirations. For this, it was considered a number of drop-out was comparatively low.

There were significantly positive correlations between the total number of participants in this program and the observational items (lip and tongue movement, pronunciation [ka], and utterance time). Namely, the frequency of the training affected the improvement of the muscle movement of the lips and the tongue (front, middle, and base) related to pronunciation and the increase in the respiratory capacity. There are some reports in the literature of sarcopenia-like changes in the muscles of the upper aerodigestive tract [19-21] and the observed age-related changes in strength and functions [5,6]. Since sarcopenia is associated with age [42-44], the muscles related to the pharyngeal stage and respiration also have risks of sarcopenia [45]. Ongoing work is underway to generate novel interventions that are effective for diminishing sarcopenia and increasing muscle strength [46]. It is necessary to set up the components of intervention menus based on pharyngeal function, strength, and the number of times of exercise per week for elderly people to acquire results that are more effective.

Furthermore, dysphagia also reduced quality of life (QOL) [39,40]. The SWAL-QOL was used to check the subjective feelings of swallowing-related QOL among the elderly. Although the score of the item "I am

Items		No dry		Almost dry		light dry		Very dry		z value	p value		
Subjective evaluation (4 point Likert scale)													
I feel dry in mouth	before	42	(57.5)	5	(6.8)	9	(12.3)	16	(21.9)	-1.239	0.215		
	after	47	(64.4)	5	(6.8)	8	(11.0)	13	(17.8)				
Subjective evaluation (3 point Likert scale)													
1.I often waked up by a fit of coughing at night.	before	63	(86.3)	7	(9.6)	3	(4.1)			-0.365	0.715		
	after	64	(87.7)	4	(5.5)	5	(6.8)						
2.I take longer time to eat.	before	49	(67.1)	6	(8.2)	18	(24.7)			-2.165	0.030*		
	after	60	(82.2)	3	(4.1)	10	(13.7)						
3.I feel sputum rolled in throat during and after eating foods or other time.	before	59	(80.8)	5	(6.8)	8	(11.0)			-1.140	0.254		
	after	65	(89.0)	3	(4.1)	5	(6.8)						
4.I choke after eating foods.	before	54	(74.0)	14	(19.2)	5	(6.8)			-0.876	0.381		
	after	58	(79.5)	11	(15.1)	4	(5.5)						
5.I cease to be able to eat hard foods.	before	42	(57.5)	11	(15.1)	20	(27.4)			-0.789	0.430		
	after	40	(54.8)	9	(12.3)	24	(32.9)						
6.I am difficult to eat stiff and dry foods because of oral dryness.	before	41	(56.2)	7	(9.6)	25	(34.2)			-1.676	0.094		
	after	50	(68.5)	4	(5.5)	19	(26.0)						
7.I remain some foods in the mouth.	before	57	(78.1)	6	(8.2)	10	(13.7)			-0.241	0.810		
	after	58	(79.5)	4	(5.5)	11	(15.1)						
8.I drop some foods from the mouth.	before	52	(71.2)	11	(15.1)	10	(13.7)			-1.380	0.167		
	after	57	(78.1)	8	(11.1)	8	(11.1)						
9.I am difficult to move a saliva and foods to the throat.	before	52	(71.2)	15	(20.5)	6	(8.2)			-0.019	0.985		
	after	52	(71.2)	15	(20.5)	6	(8.2)						
10.I choke while eating.	before	42	(57.5)	21	(28.8)	10	(13.7)			-2.090	0.037*		
	after	48	(65.8)	21	(28.8)	4	(5.5)						
11.I am troubled by sputum stick to the throat.	before	50	(68.5)	14	(19.2)	9	(12.3)			-1.613	0.107		
	after	58	(79.5)	7	(9.6)	8	(11.1)						
12.I feel discomfort and something residue in the throat when swallowing.	before	59	(80.8)	10	(13.7)	4	(5.5)			-0.059	0.953		
	after	61	(83.6)	6	(8.2)	6	(8.2)						
13.My voice became hoarse.	before	41	(56.2)	15	(20.5)	17	(23.3)			-1.604	0.109		
	after	50	(65.8)	4	(5.5)	18	(24.7)						
SWAL-QOL: Fear (5 point Likert scale)													
1.I fear I may start choking when I eat food.	before	45	(61.6)	6	(8.2)	0	(0.0)	13	(17.8)	9	(12.3)	-0.464	0.643
	after	48	(65.8)	5	(6.8)	0	(0.0)	10	(13.7)	10	(13.7)		
2.I worry about getting pneumonia.	before	45	(61.6)	7	(9.6)	0	(0.0)	8	(11.0)	13	(17.8)	-0.408	0.683
	after	44	(60.3)	7	(9.6)	0	(0.0)	14	(19.2)	7	(9.6)		
3.I am afraid of choking when I drink liquids.	before	51	(69.9)	10	(13.7)	0	(0.0)	6	(8.2)	6	(8.2)	-2.500	0.012*
	after	45	(61.6)	5	(6.8)	1	(1.4)	14	(19.2)	9	(12.3)		
4.I never noticed when I am going to choke.	before	49	(67.1)	9	(12.3)	0	(0.0)	10	(13.7)	5	(6.8)	-0.648	0.517
	after	49	(67.1)	8	(11.0)	0	(0.0)	4	(5.5)	12	(16.4)		
Total feeding (5 point Likert scale)													
I can eat with good appetite.	before	56	(76.7)	13	(17.8)	2	(2.7)	2	(2.7)			-0.737	0.461
	after	57	(78.1)	13	(17.8)	2	(2.7)	1	(1.4)				

*Analyses were performed by Wilcoxon's signed-rank test

Table 3: Comparison subjective evaluations between 2 points (n=73, n (%)).

afraid of choking when I drink liquids” worsened in the lower-ranking item “Fear” of the SWAL-QOL, the scores of the 2 items of “I take longer time to eat” and “I choke while eating” were significantly better. The results showed that some elderly may have been afraid of the aspiration, when they drink the liquid. Or the elderly people came to intentionally realize that the motion of their tongue and larynx had become better.

The contents of our new intervention, which consisted of strengthening the muscles related to the pharyngeal stages of the 5 swallowing stages, were very simple, noninvasive, and effective for use in the elderly in day care service. To date, the pharyngeal exercise

has not been included in day care service. The results of this research showed that the adoption of exercise intervention menu to prevent aspiration was effective, even for short-term intervention and by same frequency of participation. We emphasize that it is necessary to modify these kinds of exercises for the elderly to promote preventive action when eating foods and preventing aspiration or aspiration pneumonia.

Finally, this study had limited about numbers of subjects and the use of only one institution, because we avoided biases by using the same staff members and intervention methods. In the future, we would use a random controlled trial study design, identify the load and frequency

of this intervention of general rehabilitation regulation, and analyze the long-term outcome to carry out more effective exercise of pharyngeal muscles.

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Disclosures

Consent was obtained from the patient by the authors to reproduce information appearing in this article. Author(s) have provided signed confirmations to the publisher of their compliance with all applicable legal and ethical obligations in respect to declaration of conflicts of interest, funding, authorship and contributorship, and compliance with ethical requirements in respect to human test subjects. If this article contains identifiable human subject(s), author(s) were required to supply patient consent prior to publication. The author(s) declared no conflicts of interest. This study was supported by a Grant-in-Aid for Exploratory Research [Project Number: 18659678] from the Ministry of Education, Culture, Sports, Science and Technology.

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