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Original

Effects of Eggshell Supplementation on Blood Pressure in Normotensive and Mildly Hypertensive Females from Vietnam: A Match-Paired Single-Blinded Placebo-Controlled Study

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Abstract: For more than half a century, areas with high calcium contents in their water have lower rates of high blood pressure than those with lower calcium levels. To date, calcium-containing foods have not been used for hypertension prevention. Eggshell is a source of calcium, and eggshell-derived calcium has been reported to be easily absorbable. Ninety-four female participants were randomly categorized into two groups, including the control and eggshell groups. Participants received the following test diets. One type was made from eggshell containing 100-mg calcium (eggshell group) and the other type contained 100-mg cellulose (control group). Participants were fed six capsules daily for 6 months. I Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were significantly decreased in the eggshell group ($p < 0.05$). However, these values did not change in the control group ($p > 0.05$). Furthermore, the eggshell group demonstrated a significantly greater SBP and DBP reduction than the control group. These results indicated that eggshell (calcium intake of 600 mg daily) was effective in lowering SBP and DBP in older adult females from Vietnam at a high risk of hypertension.

Keywords: eggshell, blood pressure, prehypertension, Vietnamese women, intervention study

Introduction

In 1952, Jun Kobayashi of Okayama University measured the acidity and alkalinity of river water across Japan and noted that areas with acidic water had a high stroke mortality rate and those with alkaline water had a low stroke mortality rate (1). Alkaline water is hard water containing high calcium and magnesium levels. This report attracted worldwide attention, and a follow-up investigation was conducted. In 1960, Schroeder in the United States investigated the association between drinking water hardness and age-adjusted mortality rates from cardiovascular disease in 49 states(2). The results revealed that states with high drinking water hardness had lower mortality rates from cardiovascular disease, whereas states with lower drinking water hardness had higher mortality rates.

In Vietnam, stroke is the leading cause of death, accounting for 21.5% of deaths in 2019 (3). Among all hypertension-related complications, stroke is the most frequent. In Vietnam, the recommended calcium intake for adults is 1,000 mg/day (4). However, a nutritional survey conducted by Khan et al. using both his FFQ (Food Frequency Questionnaire) and 24-h recall methods reported that the average calcium intake of females from Vietnam was approximately 350 mg/day (5). LE Griffith et al. in 1999 conducted a meta-analysis of intervention studies regarding calcium intake and blood pressure (6). This review article (6) summarized studies using several calcium sources; however, it did not describe studies using eggshell as a calcium source. They identified and analyzed 42 studies, including eight studies with interventions of ≥6 months and the remaining studies with interventions

of 4–14 weeks. Calcium intake was >1,500, 1,000–1,500, and <1,000 mg/day in 17, 19, and 4 studies, respectively. This review article also encompassed studies conducted in developed countries.

Eggshell contains high levels of calcium carbonate with a porous structure (Figure 1)., whereas most other forms of calcium carbonate have a nonporous structure. Eggshell has a large surface area that comes into contact with stomach acid, enabling calcium to dissolve easily. The solubility of calcium in stomach acid is crucial for its absorption in the digestive tract. Previously, we evaluated the effects of ingesting 300 mg of calcium from eggshell per day on bone density for 12 weeks and observed that it increased bone density compared with controls (7). Simultaneously, blood pressure was lowered compared with that before ingestion; therefore, eggshell has the potential to lower blood pressure. In this study, the absence of significant differences observed in the control group may be attributed to the following reasons: some participants had normal blood pressure, the amount of calcium administered was low, and the test system was designed for improving bone density.

Therefore, this study aimed to prove the hypothesis that eggshell supplementation could exert a preventive effect in older adult females from Vietnam with normotension and mild hypertension taking a daily calcium dose of <500 mg.

In Vietnam, stroke is the leading cause of death and in 2019 (21.5%) deaths (3). Stroke is the most frequent of hypertension-related complications. The recommended amount of calcium for adults in Vietnam is 1,000 mg/day (4). However, a nutritional survey conducted by Khan et al. using both his FFQ method and his 24-hour recall method reported that the average calcium intake of Vietnamese women was approximately 350 mg/day(5). A detailed analysis of intervention studies regarding calcium intake and blood pressure can be found in a 1999 Meta-analysis by LE Griffith et al. (6). They selected/identified and analyzed 42 studies. Eight studies had interventions of 6 months or more, and the others had interventions of 4 to 14 weeks. calcium intake was >1500 mg/day in 17 studies, 1000-1500 mg/day in 19 studies, and <1000 mg/day in 4 studies.

Eggshell calcium is calcium carbonate that has a porous structure, while most other forms of calcium carbonate have a non-porous structure (Figure 1). Eggshell calcium has a large surface area that comes into contact with stomach acid, so it easily dissolves. The key to absorption of calcium from the digestive tract is its solubility in stomach acid. Previously, we evaluated the effect of ingesting 300 mg of eggshell calcium per day on bone density for 12 weeks and found that it increased bone density compared to controls (7) At the same time, blood pressure was also lowered compared to before ingestion, so eggshell calcium can be expected to have a blood pressure lowering effect. The reasons why no significant differences from the control group were observed in this study may be that the subjects included some with normal blood pressure, the amount of calcium administered was low, and the test system was designed to improve bone density.

Based on the above, this study was conducted to prove the hypothesis that eggshell calcium supplementation can be expected to have a preventive effect in elderly Vietnamese women with high risk for hypertension and a daily calcium intake less than 500 mg.

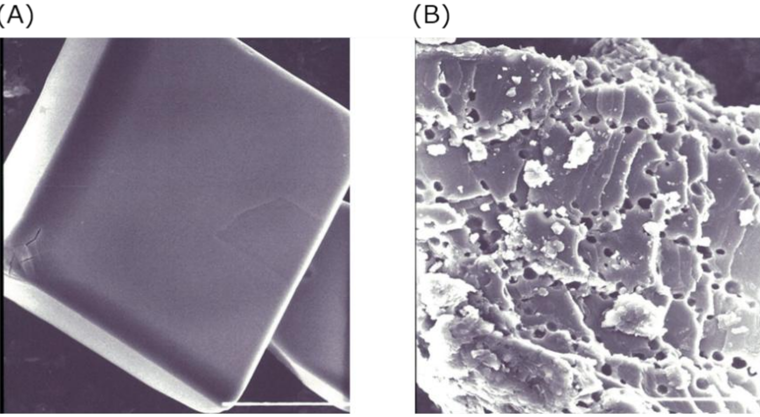


Figure 1. Microscopic photograph of eggshell calcium and calcium carbonate (8000x). A: Calcium bicarbonate B: Eggshell calcium.

Materials and Methods

Materials Calhope from Kewpie Egg Corporation. (Tokyo, Japan) was used as eggshells. Eggshells replaced with crystalline cellulose (CEOLUS FD-101; Asahi Kasei Corporation, Tokyo, Japan) were used as controls. The test food was formulated into capsules, and the composition is shown in Table 1. To support the control group, all capsules were supplemented with thiamin and riboflavin, which are not believed to influence calcium absorption or metabolism. The treatment/dose/intake was designed to provide 600 mg of calcium by consuming six capsules daily. The capsules used in the test were prepared by Aliment Industries Co., Ltd. (Shizuoka, Japan),.

Table 1. Composition of the capsules containing (mg/capsule).

Material	Egg shell Group	Control Group
Egg Shell	263 (100mg Calcium)	0
Cellulose	143	216
Thiamine hydrochloride	0.10	0.10
Riboflavin	0.10	0.10
Sucrose fatty acid ester	8.40	8.40

Subjects and test protocol We recruited 200 females aged ≥60 years. The blood pressure of each participant was measured using a manual manometer, and 120 candidates with a SBP of 120–139 mmHg or a DBP of 80–89 mmHg and were not taking any drugs were selected. Considering the potential use in disease prevention, the participants were with high-normal blood pressure and high-value blood pressure according to the 2019 Hypertension Treatment Guidelines of the Japanese Society of Hypertension (7)
The following were the rationale for selecting females: postmenopausal females are at a higher risk of heart disease, and bone density decreases owing to the effects of estrogen; therefore, the participants of this study were females with the aim that calcium supplementation would improve calcium metabolism overall. We conducted a nutritional survey on these 120 females and selected 94 females whose calcium intake was <500 mg/day)

Participants were instructed to maintain their usual diet, physical activity, sunlight exposure time, and all other activities.

The following were the inclusion criteria: [1] females aged >60 years who were postmenopausal for at least 5 years [2] blood pressure was in the prehypertension range (SBP, 120–139 mmHg; DBP, 80–89 mmHg); [3]) daily calcium intake was <500 mg/day; [4] body mass index (BMI) of 18.5–24.9 kg/m^2; and [5] agreed to participate in this study.

The following were the exclusion criteria: [1] had kidney diseases and (2) were taking other calcium-containing functional foods.

Pairs of participants matched for age, BMI, and calcium intake were randomly categorized into a control group and an eggshell group, each comprising 47 participants. Smokers and drinkers were excluded during screening.

For 6 months, participants took six capsules daily, including two capsules each after breakfast, lunch, and dinner. Height, weight, blood pressure, and bone mass were measured at the start of the study and at 6 months. Blood pressure measurements were performed at the local health center after waking up and without having breakfast at 15-min intervals. When a large difference was noted, a third measurement was taken 15 min later, and the average value of the two closest-matching results was used.

This study was conducted between April 2023 and October 2023 at Kim Thanh, Hai Duong, Vietnam. The weather conditions from April to October were almost similar.

This study complied with the guidelines of the Declaration of Helsinki and was approved by the Ethical Committee of the National Institute of Nutrition in Hanoi, Vietnam (784/VDD-QLKH). All participants provided written informed consent. This study was registered at the University Hospital Medical Information Center (UMIN; ID: UMIN000057942).

Dietary analysis This study employed the 24-h dietary recall method. Participants comprehensively recounted what they ate the day before the interview. We used the Institute of Nutrition's Album of popular dishes to help participants recall and accurately answer during the interview. The nutritional value of the diet was calculated on the basis of the Nutritional Composition Table of Vietnamese Foods 2007.

Sample size Based on the results of a previous trial wherein eggshell ingestion led to blood pressure reduction (8), the sample size was calculated using power analysis by the statistical software R ($1 - \beta = 0.80$, $\alpha = 0.05$, effect size = 0.42); 45 or more participants were required per group, and the number of participants in this study satisfied this number.

Statistical analysis Data were presented as means ± standard deviations (SDs). Comparisons of values before and after study initiation within each group were performed using the Wilcoxon signed-rank test. The Mann–Whitney U test was performed to compare both groups. Moreover, a comparison of the changes in blood pressure over a 6-month period between the two groups was performed using the Mann–Whitney U test. In all the abovementioned statistical analyses, $p < 0.05$ was considered statistically significant. All statistical analyses were performed using Statistical Package for the Social Sciences (version 20; IBM Corp., Armonk, NY, USA).

Results

Subject background Of the participants, five withdrew from both the control and eggshell groups owing to personal reasons, leaving 42 participants in the final analysis. The detailed flow diagram of the study is depicted in Figure 2. The background characteristics of the participants in both groups are presented in Table 2. Characteristics encompassed BMI, waist-to-hip ratio, SBP, DBP, and calcium intake. No significant differences were noted between the two groups ($p > 0.05$)

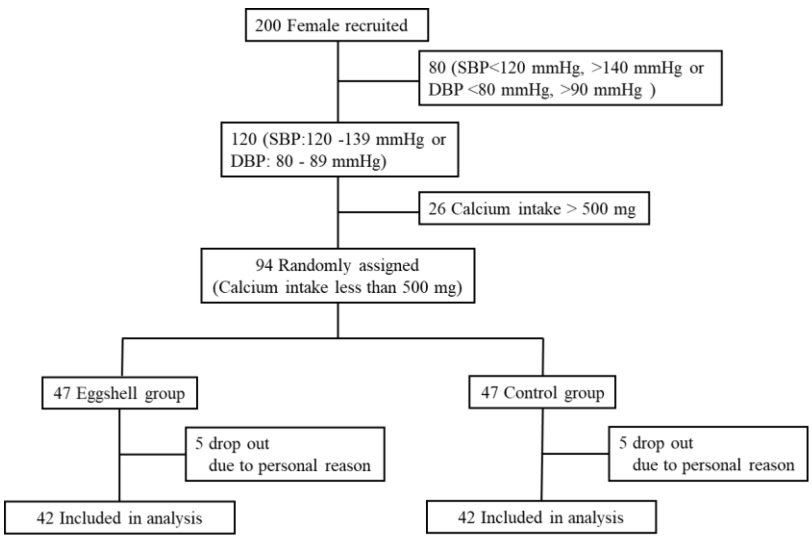


Figure 2. Participant flow through a randomized trial.

Table 2. Background characteristics of subjects between the two groups (mean±SD)

Characteristics	Eggshell Group (n=42)	Control Group (n=42)	*p* value
Age (years)	64.7 ± 3.6	65.1 ± 5.2	0.986
Weight (kg)	52.1 ± 5.7	52.4 ± 6.5	0.785
Height (cm)	152 ± 4	153 ± 5	0.704
BMI (kg/m2)	22.7 ± 2.6	22.5 ± 2.4	0.844
WHR	0.9 ± 0.1	0.9 ± 0.1	0.130
SBP (mmHg)	127 ± 7	126 ± 8	0.447
DBP (mmHg)	79.0 ± 5.9	77.0 ± 6.4	0.188
Education level			
Primary	8	8	
Secondary	29	28	
High school	5	6	
College/University	0	0	
Postgraduate	0	0	
Occupation			
Government staff /Intellectual worker	2	1	
Factory worker	1	2	
Farmer	36	35	
Trader/Business person /Self-employed in trade	1	1	
Housewife	2	3	

Dietary analysis The dietary analysis results are shown in Table 3. No significant differences in intakes of energy, protein, carbohydrates, lipids, calcium, phosphorus, sodium, potassium, and vitamins D and K1 were observed between both groups. After the test period, the bottles containing each supplement were collected from the participants, and the remaining amount was analyzed. The intake rate of the test sample was 100% across all participants.

Table 3. Energy and nutrient intakes (mean±SD)

Characteristics	Eggshell Group (n=42)	Control Group (n=42)	*p* value
Energy (Kcal)	1292 ± 433	1232 ± 447	0.499
Protein (g)	49.0 ± 22.4	47.1 ± 18.7	0.799
Carbohydrate(g)	228 ± 80	220 ± 100	0.778
Lipids (g)	20.4 ± 11.7	18.2 ± 11.6	0.258
Calcium (mg)	377 ± 254	396 ± 224	0.474
Phosphorus (mg)	642 ± 351	606 ± 253	0.932
Sodium (mg)	1369 ± 894	1217 ± 787	0.471
Potassium (mg)	1538 ± 708	1450 ± 745	0.483
Vitamin D (mg)	1.77 ± 4.08	3.47 ± 9.45	0.238
Vitamin K1 (mg)	192 ± 355	169 ± 326	0.669

Blood pressure The SBP (left chart) and DBP (right chart) at baseline and at 6 months for the eggshell and control groups are illustrated in Figure 3. SBP and DBP values significantly decreased in the eggshell group but remained unchanged in the control group.

The changes (mmHg) in SBP (left) and DBP (right) values over 6 months in the eggshell and control groups are shown in Figure 4. The eggshell group demonstrated significantly decreased SBP values compared with the control group ($p < 0.05$). Furthermore, the eggshell group exhibited significantly decreased DBP values compared with the control group ($p < 0.05$).

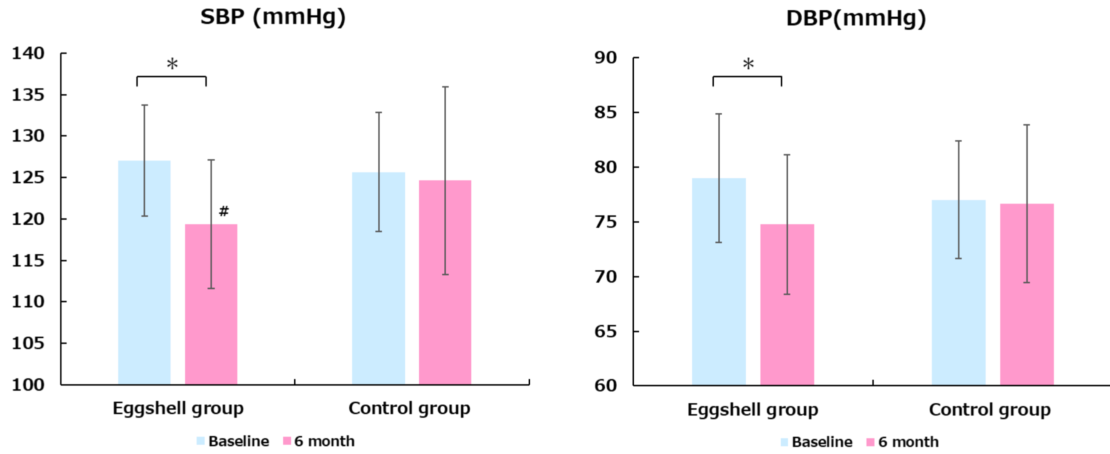
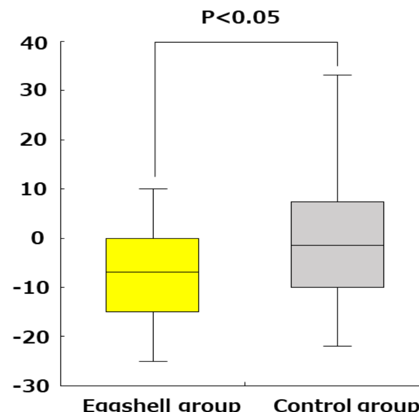


Figure 3. Changes in blood pressure after 6 months in the eggshell group and control groups (n = 42 in each group; mean ± standard deviation)
*: Significant difference compared with baseline by Wilcoxon signed-rank test ($p < 0.05$), #: Significant difference compared with the control group by the Mann–Whitney U test ($p < 0.05$)



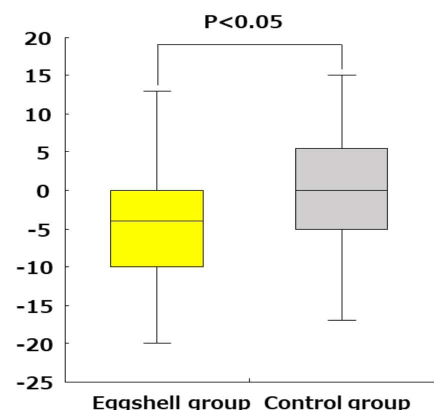


Figure 4. Boxplots comparing changes in blood pressure over 6 months. *P* <0.05 indicates a significant difference by the Mann–Whitney U test.

Discussion

We here investigated the effects of a 6-month eggshell intake on blood pressure in females from Vietnam with prehypertension (SBP of 120–139 mmHg and/or DBP of 80–89 mmHg). Two types of capsules were prepared. One contained 263 mg of eggshell (100-mg calcium) per capsule (eggshell group), and the other contained 263 mg of cellulose (control group). In the eggshell group, SBP (mmHg) significantly decreased from 127 to 119 mmHg, and DBP (mmHg) significantly decreased from 79 to 75 mmHg ($p < 0.05$). The control group did not exhibit changes in blood pressure ($p > 0.05$). In Vietnam, the recommended calcium intake for adults is 1,000 mg/day. The average daily calcium intake of the study participants was 350 mg; therefore, providing those 600 mg daily brought their intake near the recommended amount.

Here, the degrees of eggshell intake-induced blood pressure reduction were 8 and 4 mmHg for SBP and DBP, respectively. The degree of blood pressure reduction caused by materials other than calcium was as follows: sardine peptides in 13 weeks (SBP, −6.2 mmHg; DBP, −3.1 mmHg) (9) flaxseed oil in 12 weeks (SBP, −7.6 mmHg; DBP, −4.5 mmHg) (10), and milk protein tripeptides in 4 weeks (SBP, −8.0 mmHg; DBP, −3.8 mmHg) (11). Therefore, the blood pressure-lowering effect of eggshell was deemed effective, given its significant difference from the control group and the degree of reduction.

Epidemiological studies have demonstrated that a 10-mmHg reduction in SBP decreases the risk of major cardiovascular disease, coronary heart disease, stroke, and heart failure by approximately 20%, 17%, 27%, and 28%, respectively (12). In this study, an 8-mmHg reduction in SBP may reduce the risk of these diseases.

In particular, in Vietnam, stroke is the leading cause of death, accounting for 21.5% of mortalities (3); considering that calcium intake is only approximately one-third (350 mg/day) of the recommended amount (1,000 mg/day), the blood pressure-lowering effect of 600-mg/day eggshell calcium supplementation is of great public health significance. Interventions using safe and readily available food-derived ingredients, including eggshell calcium, before initiating drug therapy, may be valuable from the perspectives of reducing medical costs and avoiding drug side effects. Furthermore, as the study participants were older adult females at a high risk of osteoporosis, eggshell calcium intake may exert the dual health benefits of reducing blood pressure and improving bone density. These outcomes could contribute to extending healthy lifespan and reducing medical and nursing care-related burden in an aging society.

The following mechanism is believed to be caused by calcium intake on hypertension prevention and management. Abnormal calcium accumulation may develop in the arteries of older adults with arteriosclerosis, which initially appears to be caused by excessive calcium intake. However, this accumulation was actually due to inadequate calcium intake, known as the calcium paradox (13).

The mechanism for this is believed to be that parathyroid hormone secretion increases secondary to deficient calcium intake, releasing calcium from bones and promoting its uptake into cells (13. In other words, when calcium is insufficient, it abnormally enters the cells, keeping them constantly switched on. Muscle contraction occurs when calcium abnormally enters muscle cells due to calcium deficiency. Smooth muscles surround arteries; therefore, excessive calcium concentration causes the muscles to contract, narrowing the arteries and increasing

blood pressure. A similar process affecting the heart causes angina pectoris. The effectiveness of calcium blockers, which inhibit calcium uptake into cells, for hypertension and angina pectoris represents a good example of the strong association between calcium and cardiovascular disease. Moreover, high blood pressure can damage blood vessel walls and cause arteriosclerosis and ischemic heart disease.

To date, most studies that have investigated the effects of calcium interventions on blood pressure have been conducted in Western populations. The meta-analysis by LE Griffith et al. (6) in 1999 analyzed 42 studies. Eight studies had interventions of ≥6 months, and the remaining studies had interventions of 4–14 weeks. Calcium intake was >1,500, 1,000–1,500, and <1,000 mg/day in 17, 19, and 4 studies, respectively. Several studies have reported favorable outcomes. Moreover, a previous study in Japan demonstrated similar effects. In 2013, an epidemiological study by Umezawa et al. indicated that dietary calcium intake reduces the incidence of stroke in middle-aged individuals from Japan (14,15).

Regarding the influence of calcium on blood pressure in Asians, it is interesting to observe the association between diseases and life expectancy in Okinawa, one of Japan's 47 prefectures. Okinawa was the location of Japan's most intense ground battle during World War II. Despite its defeat in the war, Okinawans had the longest global life expectancy(16). At that time, stroke due to high blood pressure was the leading cause of mortality among individuals from Japan; however, the mortality rate for stroke among individuals from Okinawa was low. Yamamoto et al. reported that Okinawa is an island made of raised coral reefs, and the water is hard with a high calcium concentration (16). Sweet potatoes, the people's major energy source back then, and locally grown vegetables had high calcium levels. During that period, the average daily calcium intake for a Japanese adult was <500 mg, whereas for an adult in Okinawa, it was >2,000 mg (17,18) Westerners are already taking the recommended dose in Vietnam in their daily lives and are also administering 500–1,000 mg/day to achieve the desired effect. Conversely, the daily calcium intake of Vietnamese is <500 mg, which is approximately 50% that of Westerners (19). This finding suggests that even with similar additional dose is the same, 500mg, the effect of improving blood pressure can be expected to be higher in Vietnamese than in Westerners. Various calcium sources have been investigated in an attempt to prevent osteoporosis (20,21)

The eggshell is composed of 97% calcium carbonate (22,23) and has a spongy tissue structure with many small holes ((micro-pores: Figure 1). The porous eggshell structure might potentially contribute to its high solubility in the gastrointestinal tract, supporting the rapid development of organs that consume high calcium levels, including the skeleton, muscles, and brain of the chick before hatching. The calcium absorption rate of eggshell powder was 34.8%, which was higher than that of nonmicropore calcium carbonate (24). A study involving rats revealed the superiority of eggshell calcium absorption over other carbonate calcium absorption (25-27) (Figure 5). Additionally, a 3-month study involving older females receiving 2 g of calcium from eggshell and calcium carbonate reported that those who received eggshell calcium exhibited significantly suppressed parathyroid hormone secretion compared with those provided calcium carbonate (13). Furthermore, Shizuka reported that in rats, the absorption of eggshell calcium was higher than that from other carbonate calcium, and eggshell calcium consumption resulted in greater fracture strength of the femur and higher femoral calcium content than the consumption of other carbonate calcium (28). Moreover, Kikuchi et al. cited the low P content as a reason for the excellent eggshell calcium absorption (29). The ratio of calcium to P is 0.3 to 100 calcium in eggshells, and 73.3 in commercially available calcium preparations. Human health research on eggshell calcium is scarce. Hien et al. supplemented two dietary types to females from Vietnam (average age, approximately 60 years) with low bone density, which included 300 mg of eggshell calcium or 300 mg of calcium carbonate, to their daily diet (approximately 400-mg calcium/day) (8). Omi and Ezawa compared the effects of eggshell calcium supplementation with other nonmicropore carbonate calcium supplements in ovariectomized rats and observed that the eggshell calcium group had higher hip bone density than the other carbonate calcium group (30).

Although no significant difference was noted in vitamin D intake, the control group demonstrated a higher value of vitamin D intake. As an essential nutrient, vitamin D promotes calcium absorption. As the eggshell group exhibited a blood pressure-lowering effect despite having a low vitamin D intake, calcium, the main component of eggshells, may have significantly contributed to this effect.

A community health center worker with extensive experience in blood pressure taking performed all measurements using a manometer. Measurements were taken twice at 15-min intervals before breakfast; when a significant difference was noted, a third measurement was taken 15 min later, and the average value of the two similar measurements was used. Through these efforts, we believe that the data obtained were reliable. The required number of participants was approximately 60 but was increased to 94 to account for dropouts. In this study, dropouts were rare that 84 participants (42 in each group) completed the 6-month study. To reduce dropouts, researchers and community health center workers visited the participants once a week to ask about their progress and establish good relationships. The SDs of DBP and SBP changes were small (Figure 1). These findings suggest the universality of the effect of eggshell calcium on improving hypertension.

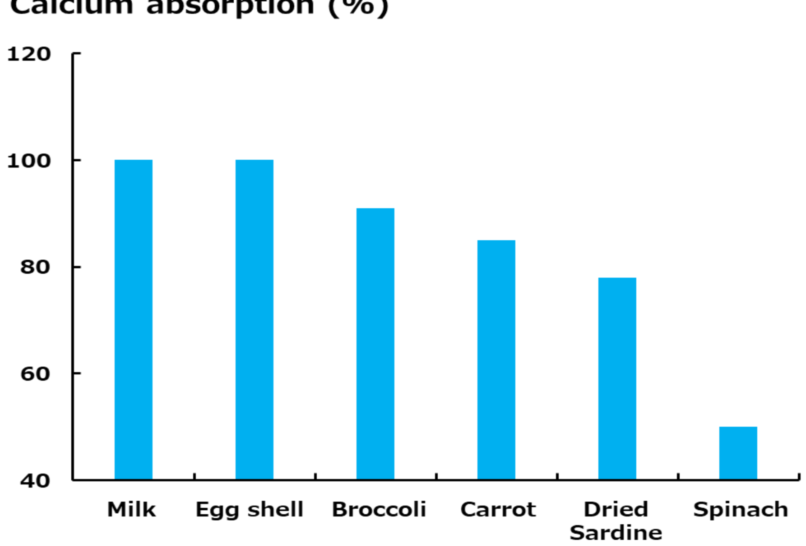


Figure 5. Calcium absorption rate of various foods when the calcium absorption rate of milk is 100% (The figure was made by extracted from the table in Reference (27).

This study had some limitations. First, it was conducted on females from Vietnam. Although the effects on males and other races were not explored, it was believed that the abovementioned mechanism would also be effective in these populations. Second, this study was conducted on untreated individuals with high blood pressure, and the effects on patients with hypertension remained unclear; so, its use in treatment is a topic for future investigation.

The large disparity between healthy life expectancy and average life expectancy has become a worldwide social problem. This gap is likely attributed to cardiovascular diseases and fractures. In this study, eggshell has been demonstrated to increase bone density but also reduces blood pressure as shown in the results of this study, so contribute to extending healthy life expectancy.

Conclusions

In conclusion, administration of Eggshell (600mg calcium/day) was effective to decrease SBP and DBP in elderly Vietnamese women with pre-hypertension.

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Original

Effectiveness of Fish Protein on Vietnamese Malnourished Type 2 Diabetes Mellitus Patients

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ABSTRACT *Background and purpose* The research was conducted to evaluate the effectiveness of the capsules made from fish protein on malnourished type 2 Diabetes Mellitus (DM) patients in Vietnam. *Methods.* A clinical controlled trial was conducted in 2 groups. Subjects in the intervention group were continuously supplemented with fish protein capsules for 12 weeks, at 9 capsules/day, while those in the control group were not. General information interview, nutritional risk assessment using the Nutritional Risk Screening (NRS) questionnaire, and nutritional status assessment using anthropometric indexes, were assessed at baseline and after 12 weeks of intervention. Serum Albumin and glucose level, as well as lipid profile conditions, and liver and kidney function were further measured before and after intervention. *Results.* 30 subjects per group met the criteria to analyze data. After 12 weeks of using fish protein capsules, the weight and nutritional status of malnourished type 2 DM patients were improved, while tests indicated the liver and kidney function were not significantly changed. *Conclusion.* The fish protein capsules improve the malnourished type 2 DM patient's nutritional status without causing side effects on liver and kidney function and is safe for human use.
Keywords: amino acids, non-toxic pufferfish, type 2 Diabetes Mellitus, malnutrition, patients, Vietnamese.

INTRODUCTION

Malnutrition (chronic energy deficiency) in adults reduces labor productivity. It increases the severity in patients with chronic diseases (cardiovascular disease, and hypertension...), increasing the rate of infectious complications leads to increased mortality (1–4). In Southeast Asia, malnutrition is estimated to account for 30-50% of adult patients in hospitals. In Vietnam, studies show that at least 1/3 of hospitalized patients are malnourished (5–7) and the rate of malnutrition in hospitalized patients is nearly 60% (8,9). Clinical characteristics of malnutrition in patients usually are unintended weight loss, decreased BMI, decreased muscle mass, and decreased fat mass. The consequences are increased risk of complications, increased severity of the disease, prolonged hospital stay, increased readmission rate, reduced quality of life, increased risk of death, and increased treatment costs (10). Therefore, planning nutritional care during hospitalization and long-term nutrition after discharge for patients is always a necessary issue (11). The Nutrition Risk Screening (NRS) nutritional risk assessment method as recommended by the European Society of Nutrition and Metabolic Diseases in 2002, is a valuable tool for evaluating the effectiveness of nutritional interventions for patients (12).

In the human body, amino acids (aa) play a prominent role, being the building blocks of proteins. Protein is an essential and vital material for the body. Protein is a component of digestive enzymes, hormones, antibodies, and serum. Therefore, when the body is not provided with enough amino acids and proteins, it will affect growth, lead to weight loss, poor digestion, and susceptibility to infections. Studies around the world have shown the effectiveness of amino acid supplementation in improving malnutrition and disease progression in patients (13–15).

Thus, it is important to find many protein and amino acid-rich sources that are available and convenient for the outpatients. Research by the Research Institute for Marine Fisheries (RIMF) have made LW-Protein Capsules from fish meat which are rich in aa and minerals. Duc Giang General Hospital belongs to Hanoi captial, Vietnam, with the number of outpatients is quite large. However, nutritional interventions at this hospital have not yet received much attention. According to Circulars of the Ministry of Health of Vietnam, patients who come for outpatient examination and treatment at the hospital also need to be screened and consulted/intervened nutrition.

Therefore, to improve the nutritional status of patients and support effective disease treatment, the research team conducted a study to evaluate the effectiveness of the fish protein capsules on nutritional status and biochemical indicators in patients with chronic energy deficiency and at risk of malnutrition coming for examination at Duc Giang General Hospital.

MATERIALS AND METHODS

1. Location: Duc Giang General Hospital, Hanoi.

2. Time: from July to December 2020.

3. Design: A clinical controlled trial was conducted in 12 weeks in 2 groups as following:

- Intervention group include patients continuously supplemented with fish protein capsules;
- Control group include patients who were not provided the capsule.

Both groups kept the same diet, and physical activity and took medications as usual.

4. Subjects and sample size

Sample size:

The sample size for each group was calcultaed as 36 patients. Then, for 2 groups, the total estimated study sample size is 72 patients. Subjects in control group was matched with intervention group by age and duration of treatment at the hospital.

Selection criteria:

Patients with type 2 DM, aged from 40 years and above, undergoing inpatient or outpatient treatment at Duc Giang General Hospital, with duration < 5 years, and no complications; without lipid disorder, BMI from 17.5 - 18.5 or NRS score ≥ 3, volunteered to participate in the research.

Exclusion criteria:

People with gastrointestinal disease or gastrointestinal surgery, cancer, heart disease, liver disease, kidney disease, or acute illness at the time of screening. People who intend to use functional foods/formula milk rich in amino acids/vitamins and minerals during the study period.

5. Methods and techniques

5.1. Screening and selecting subjects

All subjects were screened and selected by interviewing General information, nutritional risk assessment using NRS questionnaire, nutritional status assessment using BMI index.

5.2. Baseline survey (T0) and signing informed consent with the subjects

Researchers explain and invite subjects to sign informed consent to participate in the study. Assessing the patient's dietary intake by the 24-hour recall method. Fasting venous blood was collected from the patients to determine serum Albumin, Glucose, lipid profiles, GOT, GPT, and Creatinine concentration.

5.3. Providing fish protein capsules to the subjects and monitoring

The product called LW-Protein Capsules contains fish protein powder > 65%, starch and protein content > 50%, including 16 types of aa. The essential aa composition is described in detail in Table 1.

Patients in the intervention group received the dose of 9 capsules/day and used it throughout 12 weeks. Patients were guided to take 9 capsules in a day, devided into 3 times (at the time of taking breakfast, lunch and dinner). Patients also recorded their own process and illness situation in the daily report diary. After 1 week from the initial time, the researcher contacted the patient and asked about compliance and problems to provide timely instructions. Staffs from the NIN and doctors of Duc Giang General hospital were directly responsible for supervising the study during the 12-weeks of intervention period. Every month, supervisors contacted, met , and reviewed the log book to monitor capsule product intake, and adverse events.

Table 1. Composition of essential amino acids in LW-Protein Capsules.

Amino acids	In 1000 mg	In dose of 9 capsules
Isoleucine (mg)	24.98	112.41
Leucine (mg)	36.36	163.62
Lysine (mg)	44.28	199.26
Methionine (mg)	13.94	62.73
Phenylalanine (mg)	20.45	92.03
Threonine (mg)	15.19	68.36
Valine (mg)	30.82	138.67
Histidine (mg)	12.83	57.34

Patients in the intervention group received the dose of 9 capsules/day and used it throughout 12 weeks. Patients were guided to take 9 capsules in a day, devided into 3 times (at the time of taking breakfast, lunch and dinner). Patients also recorded their own process and illness situation in the daily report diary. After 1 week from the initial time, the researcher contacted the patient and asked about compliance and problems to provide timely instructions. Staffs from the NIN and doctors of Duc Giang General hospital were directly responsible for supervising the study during the 12-weeks of intervention period. Every month, supervisors contacted, met , and reviewed the log book to monitor capsule product intake, and adverse events.

6. Data analysis

Data of subjects met the following criteria were included into analysis:

• Subjects took ≥ 80% of the prescribed amount of capsules during the intervention period;

• Subjects fully participated in baseline survey and final assessments at the end of study.

All data analysis were done using Excel, STATA, and SPSS 16.0 software with statistical tests.

RESULTS

After 12 weeks of participating in the study, there were 3 subjects in intervention group and 5 subjects in control group have droped out of study. Therefore, the analysis was done in 30 pair of subjects who completed the study. Table 2 is the characteristics of the subjects at the beginning of the study (T0).

Table 2. General characteristics of the subjects at baseline

Variables	Control group (n = 30)	Intervention group (n = 30)	p*
	mean ± SD		
Age	68.5 ± 6.1	68.3 ± 5.9	> 0.05
Weight (kg)	45.3 ± 5.0	45.2 ± 6.58	
Height (cm)	152.8 ± 7.5	152.3 ± 6.9	
BMI (kg/m^2)	18.98 ± 2.2	18.95 ± 2.2	
Waist circumference (cm)	73.9 ± 5.1	74.3 ± 6.3	
Hip circumference (cm)	87.6 ± 4.8	87.3 ± 4.4	
Body fat percentage (%)	22.5 ± 8.5	22.7 ± 8.3	

T-test or Man-Whitney test

The information shows no difference in age, height, weight, waist circumference, hip circumference, and body fat percentage between the two groups at the time before the study.The results of Table 3 showed that, at baseline, there was no difference in the average concentration of biochemical indices: Albumin, Glucose; indices evaluating blood fat status including Cholesterol, Triglycerides, HDL-C, LDL-C; indices evaluating liver and kidney function: AST, ALT, Creatinine between two groups.

Table 3. Biochemical indices of the two groups at the beginning of the study

Variables	Control group (n = 30)	Intervention group (n = 30)	p*
	mean ± SD		
Albumin (g/L)	39.2 ± 2.6	39.5 ± 2.2	> 0,05
Glucose (mmol/L)	8.1 ± 3.2	9.7 ± 36	
Cholesterol (µg/L)	4.36 ± 1.24	4.44 ± 0.86	
Triglyceride (mmol/L)	1.96 ± 1.11	1.86 ± 1.22	
HDL-Cholesterol (mmol/L)	1.07 ± 0.34	1.18 ± 0.34	
LDL-Cholesterol (mmol/L)	2.55 ± 0.88	2.41 ± 0.76	
Creatinin (µmol/l)	89.7 ± 14.5	98.5 ± 11.5	
AST (U/L)	24.1 ± 11.4	28.9 ± 15.7	
ALT (U/L)	16.5 ± 15.1	17.1 ± 14.4	

T-test or Man-Whitney test

Table 4. Changes in anthropometric indexes after 12 weeks of intervention

Variables	Time	Control group (n = 30)	Intervention group (n = 30)	p*
		mean ± SD		
Waist circumference (cm)	T0	73.9 ± 5.1	74.3 ± 6.3	> 0.05
	T3	74.1 ± 5.3	74.1 ± 6.5	
	T3 – T0	0.1 ± 1.1	-0.3 ± 1.2	
Hip circumference (cm)	T0	87.6 ± 4.8	87.3 ± 4.8	> 0.05
	T3	86.9 ± 5.3	85.9 ± 3.9	
	T3 – T0	-0.8 ± 3.2	-1.1 ± 3.9	
Body fat percentage	T0	22.5 ± 8.5	22.7 ± 8.3	> 0.05
	T3	22.8 ± 8.6	23.8 ± 7.6	
	T3 – T0	0.3 ± 4.7	1.1 ± 6.3	
BMI (kg/m^2)	T0	19.0 ± 2.2	18.9 ± 2.2[a]	> 0.05
	T3	18.9 ± 2.4*	19.9 ± 2.2[a*]	< 0.05
	T3 – T0	-0.1 ± 0.2*	1.0 ± 0.6*	
Weight (kg)	T0	45.3 ± 5.0	45.2 ± 6.6[a]	> 0.05
	T3	45.2 ± 5.2*	46.4 ± 6.5[a*]	< 0.05
	T3 – T0	0.2 ± 0.4*	1.1 ± 1.6*	

**p < 0.05 comparison between 2 groups (Man-Whitney test)*
[a]p < 0.05 compare before and after intervention in the same group (paired t-test).

The results showed that after 3 months of supplementing with LW-Protein Capsules, patients had an average weight increase to 46.4 ± 6.5 (kg) and BMI of 19.9 ± 2.2, statistically significant compared to the beginning (average weight is 45.2 ± 6.6 kg and BMI is 18.9 ± 2.2). The average weight of the intervention group increased by 1.1 ± 1.6 (kg). The intervention group's waist and hip circumference indexes mildly decreased, and body fat percentage increased compared to the baseline, but there was no statistical significance in these 3 indexes. For the control group, after 12 weeks, no changes in anthropometric indexes were seen.

Regarding the effects on biochemical indexes in patients participating in the study.

Table 5. Changes in albumin concentration after 12 weeks of intervention

Variables	Time	Control group (n = 30)	Intervention group (n = 30)	p*
		mean ± SD		
Albumin (g/L)	T0	39.2 ± 2.6	39.5 ± 2.2[a]	>0.05
	T3	40.1 ± 2.7	41.1 ± 2.5[a]	<0.05
	T3 – T0	0.4 ± 0.6	1.5 ± 0.4*	

**p < 0.05 compared to the control group (Man-Whitney test)*
[a]p < 0.05 compare before and after intervention in the same group (paired t-test).

The result demonstrated that the plasma albumin concentration of the intervention group (41.1 ± 2.5 g/L) was statistically significantly higher than the control group (39.5 ± 2.2 g/L) with p<0.05. The increase in average albumin concentration before and after 3 months in the intervention group was 1.5 ± 0.4 (g/L), which was higher than the corresponding difference in the control group (0.4 ± 0.6 g/L) with p < 0.05.

Changes in blood glucose, lipid concentrations, and liver and kidney function before and after the intervention of patients are presented in Table 6.

Table 6. Changes in biochemical indexes after 12 weeks of intervention.

Variables	Time	Control group (n = 30)	Intervention group (n = 30)
		mean ± SD	
Glucose (mmol/L)	T0	8.3 ± 3.2	8.7 ± 3.6
	T3	8.4 ± 3.5	8.8 ± 4.1
	T3 – T0	0.1 ± 2.6	0.2 ± 2.4
Cholesterol (mmol/L)	T0	4.36 ± 1.24	4.44 ± 0.86
	T3	4.51 ± 1.14	4.53 ± 0.84
	T3 – T0	0.15 ± 0.85	0.09 ± 0.76
Triglycerid (mmol/L)	T0	1.96 ± 1.11	1.86 ± 1.22
	T3	1.91 ± 1.01	1.69 ± 0.97
	T3 – T0	-0.07 ± 0.68	-0.18 ± 0.87
HDL-C (mmol/L)	T0	1.19 ± 0.34	1.18 ± 0.34[a]
	T3	1.31 ± 0.36	1.34 ± 0.41[a]
	T3 – T0	0.11 ± 0.41	0.16 ± 0.20
LDL-C (mmol/L)	T0	2.55 ± 0.88	2.41 ± 0.76
	T3	2.68 ± 0.69	2.57 ± 0.79
	T3 – T0	0.12 ± 0.84	0.15 ± 0.60
Creatine (µmol/L)	T0	78.0 ± 14.1	98.45 ± 9.15
	T3	78.8 ± 14.8	97.33 ± 9.56
	T3 – T0	0.9 ± 5.4	1.12 ± 6.5
GOT (U/L)	T0	24.1 ± 11.4	28.97 ± 8.53
	T3	25.9 ± 9.0	27.93 ± 10.4
	T3 – T0	1.8 ± 4.6	1.04 ± 5.2
GPT (U/L)	T0	17.2 ± 14.6	17.1 ± 9.49
	T3	18.8 ± 9.8 [a]	18.03 ± 17.84
	T3 – T0	1.6 ± 5.3	1.02 ± 4.9

[a] *$p < 0.05$ compare before and after intervention in the same group (paired t-test).*

In the intervention group, HDL-C concentration at the end was statistically significantly higher than before the intervention. The average HDL-C concentration in the intervention group increased by 0.16 ± 0.20 mmol/L after 12 weeks of taking LW-Protein Capsules.

There was no difference in the concentrations of Glucose, Cholesterol, Triglyceride, and LDL-C between the 2 study groups before and after intervention as well as no difference when comparing between 2 time points in the same group.

The concentrations of biochemical index assessing liver and body function in both the control group and the intervention group did not change with statistical significance, proving that the product is effective in protein metabolism and patient health without causing side effects on liver and kidney function.

Table 7. Dietary intake after 12 weeks of intervention

Variables	T0 (n=36)		T3 (n=30)	
	Control	Intervention	Control	Intervention
	Median (p25; p75)		Median (p25; p75)	
Energy (kcal)	1007 (873;1417)	1078 (826;1398)	1021 (827; 1234)	1188 (1036; 1398)*
Protein (g)	40.5 (38.2; 52.4)	42.4 (28.0; 61.2)	41.3 (32.4; 48.6)	48.3 (31.7; 60.2)*
Lipids (g)	18.2 (18.9; 351)	19.6 (28.4; 38.9)	20.3 (18.5; 33.4)	27.3 (25.1; 41.0)*
Glucid (g)	176.5 (119.8; 175.3)	178.2 (121.7; 181.2)	174.7 (145.7; 206.3)	193.1 (162.2; 215.5)*
Calcium (mg)	306 (216; 474)	311 (195; 468)	289 (221; 468)	367 (272; 497)*
Iron (mg)	9.2 (5.9; 10.1)	9.1 (6.3; 9.3)	9.2 (6.1; 11.3)	10.1 (6.2; 11.6)*
Zinc (mg)	5.4 (4.4; 8.6)	5.4 (3.9; 7.6)	5.5 (4.5; 9.0)	6.3 (4.2; 8.6)*
Vitamin A (mcg)	60.1 (8.7; 151.2)	53.9 (15.6; 161.2)	60.3 (19.0; 134.3)	64.2 (12.4; 175.2)*
Vitamin C (mg)	90.5 (37.2; 153.5)	89.9 (40.8; 186.3)	91.5 (35.8; 145.1)	189.6 (99.8; 324.6)*
Folate (mcg)	193.3 (128.2; 345.8)	196.2 (116.5; 375.2)	195.8 (125.6; 367.8)	223 (135.2; 338.6)*
Vitamin D (mcg)	0.04 (0.0; 2.24)	0.03 (0.0; 2.21)	0.03 (0.0; 2.16)	0.04 (0.0; 2.37)*

**p<0.01 using the Wilcoxon rank sum-test to compare between 2 groups*

Research results showed that, at the time before intervention, there was no difference in nutrient consumption between the 2 groups. However, after 12 weeks of conducting the study, the total energy consumption, protein, lipid, carbohydrate, and micronutrients are all significantly higher in intervention group compared to the control group. The results showed that supplementing with LW-Protein Capsules after 3 months stimulated the diet of the patients, effectively increasing food consumption.

DISCUSSION

Research using capsules rich in amino acids made from fish meat on malnourished patients has shown the effectiveness of the nutritional intervention for patients with malnutrition.

The study was designed as a controlled study, evaluating both comparison with control and before - and after-intervention. The study lasted 3 months (12 weeks) in accordance with the minimum recommended duration of use of functional foods. The initial comparison showed that there was no difference between the 2 groups in terms of basic information assessing nutritional status, biochemical indices assessing protein intake, blood sugar status, lipid profile, liver, and kidney function, showing that 2 groups and sample sizes were appropriate to conduct the study (Tables 1, 2, 3). The average BMI of all subjects was about 19.0, in the normal range (because the study used BMI criteria from 17.5 - 18.5 for people 40 - 70 years old, or BMI < 20 for people 71 - 80 years old) but is the lower limit of normal, indicating the patient's malnutrition. After 12 weeks of intervention research, the 2 groups had the above indicators measured again and compared. The results showed an increase in the patient's BMI and weight (Table 4), demonstrating an improvement in the patient's weight. In addition, albumin concentration, representing protein in the patient's body, also increased with statistical significance. Besides, to further understand the nutrient consumption of participants, diets were also analyzed, before and after using the product (Table 7). The results showed that there was a significant improvement in the nutritional diet of patients using the product.

However, the improvement results are not too great, possibly because the time of using the product is not long, and the patient's psychological condition affects eating and absorption. The study also did not evaluate the effectiveness of the LW-Protein Capsules on the improvement of the disease progression. However, the research results have resolved some of the issues mentioned above, showing the safety of the product, and the role of nutritional intervention (essential amino acids) on humans (patients).

In conclusion, after 12 weeks of using LW-Protein Capsules, the weight and nutritional status of diabetic patients being treated at Duc Giang General Hospital were improved, while the liver and kidney function of the

subjects were not increased, proving that the product is effective for the patient's nutritional health without causing side effects on liver and kidney function, and the product is safe for users.

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Original

Associations of Appetite with Anthropometric Measurements, Mental Health, Dietary and Nutritional Status: A Cross-Sectional Study in Elderly Japanese People Attending A Day-Care Facility

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ABSTRACT Our study investigated the prevalence of poor appetite (PA) and its associations with anthropometric measurements, mental health, dietary and nutritional status in the elderly. We analyzed 71 participants (26 men, 45 women) at a day-care facility in City N and surveyed their basic characteristics (age, sex, support/care level, and living status), anthropometric measurements (including height, weight, calf circumference), mental health status (Geriatric Depression Scale-15 [GDS-15], Philadelphia Geriatric Center Morale Scale [PGC-MS]), appetite (Japanese version of the Council on Nutrition Appetite Questionnaire), and nutritional status (Mini Nutritional Assessment, food frequency questionnaire). The participants were divided into a PA group and a good appetite (GA) group. Between-group differences were examined using the Mann-Whitney U test or chi-square test. Binary logistic regression analysis was conducted assigning 0 to PA and 1 to GA as dependent variables and using the relevant factors as independent variables. The prevalence of PA was 38% overall. Significant between-group differences were noted for living status, calf circumference, GDS-15 score, and PGC-MS. Intakes of energy, protein, other nutrients and various food groups in the PA group were significantly lower than those in the GA group. The proportion of individuals with insufficient/inadequate intake of energy was greater in the PA group than in the GA group. Regression analysis demonstrated significant relationships of appetite with living status (odds ratio 3.838, 95% confidence interval 1.136–12.969) and protein consumption/standard body weight (9.449, 1.285–69.487). Assessment of appetite in elderly persons needing nursing support/care seems important for the prevention of undernutrition or malnutrition.

Keywords: appetite, assessment, elderly, nutrient consumption, undernutrition

INTRODUCTION

Undernutrition or malnutrition constitutes a major health problem in the elderly, causing sarcopenia and frailty, increasing the need for nursing support/care, and acting as a trigger for higher morbidity and mortality (1-3). Screening with the Mini Nutritional Assessment (MNA®) tool found that approximately 50% of community-dwelling elderly persons attending day-care facilities or receiving home care were at risk of malnutrition and 10%–20% were malnourished (4-6); these figures were 40%–60% and 15%–30%, respectively, for elderly persons residing in nursing homes (7-9). Thus, the risk of malnutrition can be as high as 50% in elderly persons, whether they live in the community or in nursing homes.

Poor appetite (PA) appears to precede weight loss and undernutrition by causing lowered and/or imbalanced intake of energy and nutrients. Appetite is affected by various factors in the elderly, including age-related frailty and chronic diseases, socioeconomic factors including financial difficulty, residential and living status, and psychological stress (10,11). To the best of our knowledge, there have been few surveys of the associations of appetite with these factors, dietary and nutritional status in elderly persons attending day-care facilities in Japan. In this study, we investigated the prevalence of PA and associations of appetite with anthropometric measurements, mental health, dietary and nutritional status in an elderly Japanese population for early intervention to prevent undernutrition or malnutrition.

MATERIALS AND METHODS

Study population. Eighty-three elderly individuals attending a day-care facility in City N, Aichi Prefecture, Japan, were initially eligible for an interview-based survey conducted between February and March 2015. After excluding 12 individuals due to incomplete information, data from the remaining 71 participants (26 men, 45 women) were available for analysis. The study protocol was approved by the Ethics Committee of Nagoya University of Arts and Sciences (approval number 94), and all participants provided written informed consent.

Basic characteristics. We obtained information on age, sex, care needs level (Support level 1 or 2, Care level 1–4) as defined by the Long-term Care Insurance System (12), number of medicines (categorized as 0 for fewer than 4 prescribed agents, and 1 for 4 or more agents), and living status (living alone or with others).

Anthropometric measurements. Height was measured using a digital height measurement device (YG-200DN; Yagami KK, Osaka, Japan). Body weight and body composition were determined using a portable device (In Body 430; Takumi KK, Tokyo, Japan). BMI was calculated as weight (kg)/height (m)2. Grip strength was measured twice in the left hand and right hand each using a digital grip dynamometer (T.K.K. 5401; Takei Instrument Industry KK, Niigata, Japan), and the average values were recorded. Arm and calf circumferences were measured, and the average values were calculated for both sides.

Mental health. Depression was assessed using the Geriatric Depression Scale (GDS-15) (13,14). A negative answer was scored as 1 and a positive answer as 0, with a maximum total score of 15. A total score of 0–4 indicates no depression, a total score of 5–10 a tendency for depression, and a total score of ≥11 depression. In this study, participants with a GDS-15 score of <5 were considered to be non-depressed, while those with a score of ≥5 to be depressed.

The Philadelphia Geriatric Center Morale Scale (PGC-MS) was used to measure subjective psychosocial well-being (15). An affirmative answer was scored as 1 and a negative answer as 0, with a maximum total score of 17. According to the instrument guidelines, scores of 13–17 were considered high, scores of 10–12 within the mid-range, and scores <9 at the low end of the scale.

Appetite. Appetite was assessed using the Japanese version of the Council on Nutrition Appetite Questionnaire (CNAQ-J) (16). The original version was developed by Wilson et al. (17). This questionnaire comprises 8 items with a maximum of 5 points each, for a total of 40 points. Individuals with a score of 8–16 may be at risk of anorexia and need nutritional counseling, those with a score of 17–28 may need frequent re-evaluation, and a score ≤28 may

predict at risk of 5% weight loss within 6 months. In our study, individuals with a CNAQ-J score of ≤28 were allocated to a PA group and those with a score of ≥29 to a good appetite (GA) group.

Dietary and nutritional status. Dietary and nutritional status was evaluated using the MNA® (18), which consists of 6 screening items (maximum 14 points) including changes in food intake and body weight, and 12 assessment items (maximum 16 points) such as living environment and prescribed medication status. Scores were summed (maximum 30 points), and nutritional status was defined as malnutrition <17 points, at risk of malnutrition 17–23.5 points, or normal nutrition ≥24 points.

Participants were also assessed using a food frequency questionnaire with confirmed validity and reproducibility (version 3.5, based on the Standard Tables of Food Composition in Japan) (19), which asks about the frequency of intake of 29 food items with 10 cooking methods. Intakes were estimated for energy, 4 macronutrients (protein, lipids, carbohydrates, and dietary fibers), 8 minerals and vitamins (calcium, iron, zinc, vitamins A, B1, B2, C, and D) and were calculated for 13 food groups (cereals, potatoes, legumes, green and yellow vegetables, other vegetables, seaweeds, fish and shellfish, meat, eggs, milk and dairy products, fruits, confectionery, oils and fats).

Intakes of energy and protein per standard body weight (kg) (22.0 [BMI] × height [m]2) were also calculated in addition to crude values and per body weight values (kg). BMI <21.5 was considered to reflect insufficient consumption of energy according to the 2020 Dietary Reference Intakes for Japanese (20).

Statistical analysis. Median differences in participant's characteristics, anthropometric measurements, mental health, and nutritional status were compared between the PA and GA groups using the Mann-Whitney U test, a 2×2 table examined by the chi-square test or Fisher's direct method, or a 2×3 table examined by the chi-square test. Spearman's rank correlation coefficients were calculated between CNAQ-J and relevant parameters. Using the forced entry method, binary logistic regression analysis was performed assigning 0 to PA and 1 to GA as the dependent variables, with relevant factors as independent variables adjusted for age and sex. In view of the rather small sample size, the independent variables were narrowed down considering the correlation coefficients and collinearity of the variables in the categories of mental health and nutrient intake. All statistical analyses were conducted using SPSS version 22.0 (IBM Corp., Armonk, NY). A two-sided p-value of <0.05 was considered statistically significant.

RESULTS

Basic characteristics. The 71 participants (26 men [36.6%], 45 women [63.4%]) had a median age of 82 years (interquartile range [IQR] 76, 87) (Table 1). The prevalence of PA was 38.0% (n=27) and that of GA 62.0% (n=44). The CNAQ-J scores were 29 (IQR 27, 32) for all participants, 27 (IQR 20, 28) for the PA group, and 31 (IQR 30, 33) for the GA group.

Overall, 30 participants (42.3%) required Support level 1 or 2, 32 (45.1%) Care level 1 or 2, and 9 (12.7%) Care level 3 or 4. There was no significant difference in age, sex, Support/Care level, or number of prescribed agents between the PA and GA groups. The prevalence of living alone was significantly greater in the PA group than in the GA group (51.9% vs 27.3%, p=0.045).

Height could not be measured in 3 participants and body weight could not be measured in 10 because of difficulty in standing erect, having a rounded spine, or declining to participate in the examination. Therefore, the analyses of nutrient intake per standard body weight were based on data for 68 participants, while those involving body weight, BMI, and skeletal muscle mass were based on data for 61 participants. BMI was lower in the PA group than in the GA group (21.0 [IQR 19.5, 23.6] vs 22.8 [IQR 20.1, 25.7]); however, the difference was not statistically significant. Calf circumference was significantly smaller in the PA group (31.0 cm [IQR 29.0, 33.0] vs 33.0 cm (IQR 30.3, 35.0]; p=0.033). There was no significant between-group difference in skeletal muscle mass, arm circumference, or grip strength.

Mental health. The GDS-15 score was significantly higher in the PA group than in the GA group (5 [IQR 3, 8] vs 3 [IQR 1, 6]; p=0.005). The proportion of depression was greater in the PA group (55.6% vs 31.8%), but the difference was not statistically significant (p=0.081). The PGC-MS score was significantly lower in the PA group (10 [IQR 7, 12] vs 12 [IQR 9, 13]; p=0.040).

Table 1. Characteristics of the elderly participants, and comparison of anthropometric measurements and mental health by appetite status

	All participants		Poor appetite group		Good appetite group		*p*-value [b]
	n	Median [IQR [a]] or (%)	n	Median [IQR] or (%)	n	Median [IQR] or (%)	
CNAQ-J score [c]	71	29 [27, 32]	27	27 [20, 28]	44	31 [30, 33]	<0.001
Age (years)	71	82 [76, 87]	27	81 [74, 87]	44	82 [77, 80]	0.614
Sex (men/women)	26/45	(36.6)	10/17	(37.0)	16/28	(36.4)	0.999
LTCI care needs level [d]							
Support level 1 or 2	30	(42.3)	18	(40.7)	19	(43.2)	
Care level 1 or 2	32	(45.1)	14	(51.8)	18	(40.9)	0.927
Care level 3 or 4	9	(12.7)	2	(7.4)	7	(15.9)	
At least 4 prescribed medicines	45	(63.4)	18	(66.7)	27	(61.4)	0.801
Living alone	26	(36.6)	14	(51.9)	12	(27.3)	0.045
Anthropometric measurements							
Height (cm)	68	148.7 [142.9, 154.7]	27	149.3 [141.5, 156.2]	41	147.8 [143.0, 154.4]	0.764
Body weight (kg)	61	49.9 [42.9, 56.7]	24	46.9 [41.1, 55.5]	37	52.6 [43.5, 57.7]	0.238
BMI (kg/m²)	61	22.1 [19.8, 25.3]	24	21.0 [19.5, 23.6]	37	22.8 [20.1, 25.7]	0.112
Skeletal muscle mass (kg)	61	17.4 [15.2, 20.8]	24	16.7 [15.4, 19.5]	37	17.6 [15.0, 21.0]	0.585
Arm circumference (cm)	71	26.0 [23.0, 28.0]	27	26.0 [22.0, 28.0]	44	27.0 [24.0, 28.0]	0.225
Calf circumference (cm)	71	32.0 [30.0, 34.0]	27	31.0 [29.0, 33.0]	44	33.0 [30.3, 35.0]	0.033
Grip strength (kg)	71	17.0 [12.9, 21.3]	27	16.6 [12.9, 19.2]	44	17.9 [12.9, 22.0]	0.485
Mental health							
GDS-15 score [e]	71	4 [2, 6]	27	5 [3, 8]	44	3 [1, 6]	0.005
Depression (GDS-15 score ≥ 5)	29/71	(40.8)	15/27	(55.6)	14/44	(31.8)	0.081
PGC Morale Scale score [f]	71	11 [8, 13]	27	10 [7, 12]	44	12 [9, 13]	0.040

[a] Interquartile range.

[b] Median values were examined by the Mann-Whitney *U* test, and proportions by the chi-square test or Fisher's direct test.

[c] Council on Nutrition Appetite Questionnaire for Japanese.

[d] Long-Term Care Insurance System.

[e] Geriatric Depression Scale-15.

[f] Philadelphia Geriatric Center Morale Scale.

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Dietary and nutritional status. The MNA® score was 24 (IQR 22, 26) and the proportion with malnutrition or at risk of malnutrition was 49.2% overall with no significant difference between the GA and PA groups (Table 2).Overall, dietary intakes were as follows: energy 1,473 kcal (IQR 1,228, 1,703), protein 52.8 g (IQR 43.5, 65.5), lipids 42.5 g (IQR 33.0, 55.2), carbohydrates 212.6 g (IQR 180.1, 236.1), and zinc, a crucial mineral for taste that influences appetite, 6.6 mg (IQR 5.4, 7.6). Intakes of nutrients (energy, protein, lipids, carbohydrates, dietary fibers, iron, zinc, vitamins B_1, B_2, C, and D) were significantly lower in the PA group than in the GA group (p<0.05)

No significant difference was noted in energy consumption per body weight between the two groups; however, energy consumption per standard body weight was significantly lower in the PA group than in the GA group (27 kcal/kg [IQR 21, 35] vs 33 kcal/kg [IQR 28, 36]; *p*=0.008). The proportion of individuals with insufficient energy consumption (BMI<21.5kg/m²) was greater in the PA group (62.5% vs 32.4%; *p*=0.034).

No significant difference was noted in protein consumption per body weight between the two groups, either; however, the value per standard body weight was significantly lower in the PA group than in the GA group (0.97 g/kg [IQR 0.73, 1.20] vs 1.19 g/kg [IQR 0.99, 1.46]; *p*=0.003).

Table 2. Comparison of nutritional status and intakes of nutrients and food groups according to appetite status

	All participants		Poor appetite group		Good appetite group		*p*-value [b]
	n	Median [IQR [a]] or (%)	n	Median [IQR] or (%)	n	Median [IQR] or (%)	
Nutritional status							
MNA® score [c]	61	24 [22, 26]	24	24 [20, 26]	37	25 [22, 26]	0.103
Malnutrition or at risk of malnutrition (MNA® score ≤23.5)	30	(49.2)	13/24	(54.2)	17/37	(45.9)	0.605
Consumption of energy and nutrients							
Energy (kcal)	71	1,473 [1,228 1,703]	27	1,330 [1,107, 1,578]	44	1,605 [1,334, 1,745]	0.006
Energy (kcal)/body weight (kg)	61	30 [25, 35]	24	29 [23, 35]	37	31 [26, 36]	0.281
Energy (kcal)/standard body weight (kg) [d]	68	30 [25, 35]	27	27 [21, 35]	41	33 [28, 36]	0.008
Insufficient energy intake (BMI<21.5)	27/61	(44.3)	15/24	(62.5)	12/37	(32.4)	0.034
Protein (g)	71	52.8 [43.5, 65.5]	27	45.8 [33.6, 56.0]	44	55.8 [49.6, 71.3]	0.003
Protein (g)/body weight (kg)	61	1.08 [0.85, 1.32]	24	0.98 [0.75, 1.23]	37	1.18 [0.92, 1.33]	0.057
Protein (g)/standard body weight (kg)	68	1.03 [0.84, 1.37]	27	0.97 [0.73, 1.20]	41	1.19 [0.99, 1.46]	0.003
Lipids (g)	71	42.5 [33.0, 55.2]	27	38.6 [26.1, 46.4]	44	46.4 [34.7, 60.3]	0.015
Carbohydrates (g)	71	212.6 [180.1, 236.1]	27	198.1 [149.4, 226.2]	44	217.0 [195.0, 245.1]	0.040
Dietary fibers (g)	71	11.6 [8.8, 13.7]	27	9.6 [7.1, 12.2]	44	12.6 [9.4, 14.9]	0.004
Calcium (mg)	71	477 [331, 554]	27	426 [282, 525]	44	488 [412, 597]	0.061
Iron (mg)	71	5.9 [4.5, 7.3]	27	5.6 [3.9, 6.3]	44	6.0 [4.8, 7.9]	0.017
Zinc (mg)	71	6.6 [5.4, 7.6]	27	5.8 [4.1, 6.9]	44	7.0 [5.9, 8.4]	0.002
Vitamin A (µg RAE) [e]	71	493 [330 , 629]	27	438 [322, 523]	44	540 [351, 663]	0.079
Vitamin B_1 (mg)	71	0.73 [0.56, 0.88]	27	0.66 [0.53, 0.78]	44	0.79 [0.58, 1.00]	0.025
Vitamin B_2 (mg)	71	0.84 [0.66, 1.04]	27	0.73 [0.61, 0.89]	44	0.93 [0.71, 1.12]	0.025
Vitamin C (mg)	71	96 [64, 116]	27	71 [44, 99]	44	103 [71, 124]	0.003
Vitamin D (µg)	71	6.4 [3.6, 8.1]	27	4.2 [2.9, 7.0]	44	6.7 [4.7, 8.9]	0.010
Consumption of food groups							
Cereals (g)	71	336 [270, 398]	27	330 [222, 384]	44	345 [314, 405]	0.135
Potatoes (g)	71	21 [7, 43]	27	14 [7, 29]	44	21 [7, 50]	0.144
Green and yellow vegetables (g)	71	75 [50, 111]	27	71 [50, 100]	44	86 [50, 123]	0.149
Other vegetables (g)	71	129 [95, 193]	27	109 [91, 160]	44	167 [104, 210]	0.023
Seaweeds (g)	71	3 [1, 5]	27	2 [1, 4]	44	5 [2, 7]	0.003
Legumes (g)	71	35 [20, 55]	27	20 [10, 40]	44	40 [30, 69]	0.002
Fish and shellfish (g)	71	54 [31, 79]	27	41 [23, 70]	44	68 [45, 91]	0.006
Meat (g)	71	51 [29, 80]	27	49 [20, 63]	44	69 [34, 91]	0.060
Eggs (g)	71	14 [7, 29]	27	14 [7, 25]	44	21 [14, 36]	0.142
Milk and dairy products (g)	71	92 [33, 190]	27	109 [8, 186]	44	87 [50, 195]	0.648
Fruits (g)	71	86 [43, 150]	27	75 [32, 107]	44	150 [64, 150]	0.021
Confectionery (g)	71	27 [10, 52]	27	43 [11, 65]	44	23 [9, 47]	0.213
Fats and oils (g)	71	6 [3, 11]	27	5 [2, 9]	44	8 [4, 13]	0.021

[a] Interquartile range.

[b] Median values were examined using the Mann-Whitney *U* test, and proportions by the chi-square test.

[c] Mini Nutritional Assessment.

[d] Standard body weight: [22×height(m)^2] kg.

[e] Retinol activity equivalents (µg).

Correlations between CNAQ-J score and relevant factors. The Spearman's rank correlation coefficient between the CNAQ-J score and calf circumference was statistically significant (r=0.313, p=0.008), as was the PGC-MS score (r=0.313, p=0.008). (Table 3). The GDS-15 score was negatively associated with CNAQ-J score (r=
-0.313, p=0.008). The correlation coefficients between the CNAQ-J score and age, BMI, and MNA® were not statistically significant.

Intakes of energy (*r*=0.345, *p*=0.003), protein (*r*=0.370, *p*=0.001), dietary fibers (*r*=0.329, *p*=0.005), zinc (*r*=0.395, *p*=0.001), and other 8 nutrients were significantly associated with CNAQ-J score. Intakes of legumes (*r*=0.332, *p*=0.005), fish and shellfish (*r*=0.309, *p*=0.009), and other 4 food items were also significantly associated with CNAQ-J score.

Table 3. Correlations between CNAQ-J[a] score and anthropometric measurements, mental health status and dietary consumption

	n	*r* [b]	*p* -value
Age (years)	71	0.033	0.783
BMI	61	0.230	0.074
Calf circumference (cm)	71	0.313	0.008
GDS-15 score [c]	71	-0.313	0.008
PGC-MS [d]	71	0.313	0.008
MNA® score [e]	61	0.231	0.073
Energy (kcal)	71	0.345	0.003
Energy (kcal)/body weight (kg)	61	0.229	0.076
Energy (kcal)/standard body weight (kg) [f]	68	0.375	0.002
Protein (g)	71	0.370	0.001
Protein (g)/body weight (kg)	61	0.299	0.019
Protein (g)/standard body weight (kg)	68	0.405	0.001
Lipids (g)	71	0.288	0.015
Carbohydrates (g)	71	0.276	0.020
Dietary fibers (g)	71	0.329	0.005
Calcium (mg)	71	0.265	0.025
Iron (mg)	71	0.281	0.018
Zinc (mg)	71	0.395	0.001
Vitamin A (μg RAE) [g]	71	0.231	0.052
Vitamin B_1 (mg)	71	0.277	0.019
Vitamin B_2 (mg)	71	0.308	0.009
Vitamin C (mg)	71	0.348	0.003
Vitamin D (μg)	71	0.272	0.022
Cereals (g)	71	0.200	0.094
Potatoes (g)	71	0.144	0.232
Green and yellow vegetables (g)	71	0.167	0.163
Other vegetables (g)	71	0.260	0.029
Seaweeds (g)	71	0.372	0.001
Legumes (g)	71	0.332	0.005
Fish and shellfish (g)	71	0.309	0.009
Meat (g)	71	0.187	0.121
Eggs (g)	71	0.233	0.050
Milk and dairy products (g)	71	0.144	0.231
Fruits (g)	71	0.300	0.011
Confectionery (g)	71	-0.130	0.280
Fats and oils (g)	71	0.270	0.023

[a] Council on Nutrition Appetite Questionnaire for Japanese.
[b] Spearman's rank correlation coefficient.
[c] Geriatric Depression Scale-15.
[d] Philadelphia Geriatric Center Morale Scale.
[e] Mini Nutritional Assessment.
[f] Standard body weight: [22 × height (m)^2] kg.
[g] Retinol activity equivalents (μg).

Binary logistic regression analysis. The GDS-15 score was negatively associated with the PGC-MS score (r= -0.623), and protein consumption was strongly correlated with intakes of energy (r=0.907) and zinc (r=0.962). Calf circumference, living status, GDS-15 score (as a mental health status marker), and protein consumption (as a marker of energy and nutrient intake) were used as independent variables. Analyses were performed using two models (model 1, crude protein consumption; model 2, protein consumption per standard body weight). Factors associated with appetite were living status (odds ratio [OR] 4.237, 95% confidence interval [CI] 1.246–14.408) and crude protein consumption (OR 1.050, 95% CI 1.007–1.095) according to model 1, while living status (OR 3.838, 95% CI 1.136–12.969) and protein consumption per standard body weight (OR 9.449, 95% CI 1.285–69.487) according to model 2 (Table 4).

Table 4. Final results of binary logistic regression analysis according to the forced entry method

	Model 1[a]				Model 2[b]			
	OR[c]	95% CI[d]		*p*-value	OR	95% CI		*p*-value
Calf circumference (cm)	1.116	0.911	1.367	0.291	1.095	0.893	1.344	0.381
Living status[e]	4.237	1.246	14.408	0.021	3.838	1.136	12.969	0.030
GDS-15 (score)[f]	0.850	0.710	1.017	0.076	0.853	0.711	1.024	0.089
Crude protein consumption (g)	1.050	1.007	1.095	0.023	–	–	–	–
Protein consumption (g)/standard body weight (kg)[g]	–	–	–	–	9.449	1.285	69.487	0.027

[a] Model 1: calf circumference, living status, GDS-15, and crude protein consumption.

[b] Model 2: calf circumference, living status, GDS-15, and protein consumption/standard body weight.

[c] Odds ratio: adjusted for sex and age.

[d] 95% confidence interval.

[e] 0, living alone; 1, living with other(s).

[f] Geriatric Depression Scale-15.

[g] Standard body weight: [22 × height (m)^2] kg.

DISCUSSION

Prevalence of PA. In previous studies, the proportion of community-dwelling elderly with PA was 10%–25% when evaluated using the CNAQ or the Simplified Nutritional Appetite Questionnaire (SNAQ), while that of individuals in need of nursing support/care, outpatients, or hospitalized patients was 20%–45% (21-24). The prevalence of PA was 38.0% in this population. These figures suggest that elderly people who need nursing support and/or medical care are at higher risk of PA.

Anthropometric characteristics. Some studies have demonstrated associations of appetite with age and several anthropometric parameters, including BMI, skeletal muscle mass, calf circumference, and grip strength, in the elderly (25-27); however, the observed associations were not universal or consistent. In this study, we found a significant association of appetite with only calf circumference. These inconsistencies may be due in part to the different backgrounds of the study populations and the fact that physical changes occur as a result of persistent loss of appetite.

Living status. Similar to the present study, there have been some studies examining associations between eating alone and/or living alone and decreased appetite (27,28). Other studies have demonstrated that living alone and/or eating alone was associated with reduced consumption of energy, nutrients and certain food items and with malnutrition (29-31). Thus, living alone and/or eating alone can be considered a risk factor for PA or malnutrition in the elderly. The prevalence of living alone (36.6%) was greater in our participants than in the

general population. Approximately 6.24 million people (26.3%) aged 65 years or over were living alone in Japan in 2015 (32). Given that elderly persons living alone have been increasing both in number and in proportion, careful attention should be paid to their appetite.

Mental health. The prevalence of depression of approximately 41% in the present study was greater than the range of 10%–30% previously reported for the elderly without functional impairment (21,33,34) but similar to the range of 30%–65% reported in the elderly living in nursing homes (7,21,35), suggesting that elderly people who need nursing support/care are at higher risk of depression. Relevant studies have found that depression was significantly related to PA/anorexia and malnutrition in the elderly living in the community and in nursing homes (7,21,34), as seen in the present study.

Although some studies have demonstrated an association of PA/anorexia with QOL in patients with cancer (36-38), there is limited information on this relationship in the community-dwelling elderly or those who need nursing support/care. Acar Tek *et al.* reported that appetite was associated with mental and physical components of health-related QOL (39). There were no published observations on the relationship between the PGC-MS score and appetite in the elderly who need nursing support/care, whereas we noted that PGC-MS score was lower in the PA group than in the GA group. Therefore, it seems worthwhile to further investigate associations between PA/anorexia and QOL.

Dietary and nutritional status. Using the SNAQ and a 24-hour dietary recall survey, Hara *et al.* demonstrated significantly lower intakes of energy, protein, lipids, carbohydrates, dietary fibers, iron, and zinc, but not of calcium or sodium, in elderly outpatients with PA/anorexia compared with their counterparts without PA/anorexia in Brazil (24). van der Meij *et al.* administered a food frequency questionnaire to a group of community-dwelling elderly in the Netherlands and found lower intakes of protein and dietary fibers in individuals with PA than in those with GA/very GA (40). Payette *et al.* also reported associations of appetite with intakes of energy and protein in an elderly Canadian population receiving publicly funded home care packages (41).

Hara *et al.* reported that outpatients with anorexia had inadequate intakes of 87.7% for energy and 71.5% for protein: that is, they were below 30 kcal/kg (body weight) for energy and below 1 g/kg for protein according to the values recommended in the European Society for Clinical Nutrition and Metabolism guideline (24,42). Payette *et al.* found that 78% of elderly men and 70% of women had insufficient energy intakes, and 57% of elderly men and 80% of women did not meet the recommended protein consumption per day (0.8 g/kg) according to the advised nutrient intake for Canadians (41,43). In the present study, 62.5% of the PA group had insufficient energy consumption as defined by the Dietary Reference Intakes for Japanese. Median values for protein consumption per body weight and standard body weight in the PA group were under 1 g/kg, while those in the GA groups were over 1 g/kg, suggesting more than half of the PA group had insufficient protein intake. Given that the participants in the above-mentioned studies (24,40,41) had varied background characteristics and different criteria were used to determine insufficient dietary intake, a direct inter-comparison could not be made; however, it seemed obvious that participants with PA had lower/inadequate consumption of energy and protein than those with GA.

In Italy, Donini *et al.* reported that elderly participants with anorexia (in community-dwelling individuals aged ≥65 years, nursing home residents, patients in rehabilitation and emergency wards) had reduced intake in certain food groups including meat, eggs, fish, fruits and vegetables (44). van der Meij *et al.* also reported that their participants with PA had significantly decreased consumption of solid food, protein-rich food, whole grains, fruits and vegetables, but increased consumption of dairy food, oils and fats, sweets, and sodas when compared with those with GA (40). Our study also noted that PA was associated with lower or inadequate intake of various food items and nutrients in elderly population attending a day-care facility. All the aforementioned studies demonstrated that PA was strongly related to reduced intakes of crucial nutrients (including energy and protein) and relevant food items. In other words, it appeared that elderly people with PA were at risk of undernutrition or malnutrition.

Binary logistic regression. A multivariate binary logistic regression noted that PA was associated with reduced consumption of protein and living alone in the present study. We also detected a strong correlation between intakes of energy and protein (r=0.907). These results

were compatible with the findings by Payette *et al.* (41), showing an association between decreased energy consumption and PA. Accordingly, consumption of nutrients (energy and protein, in particular) and living status seemed important factors of PA in elderly people.

Limitations. The main limitation of this study was its lack of statistical power because of a small study population. It had a cross-sectional design, which precluded investigation of cause-and-effect relationships and the risk of PA. Furthermore, because this study was conducted in a day-care facility, it appears unclear whether the findings could be generalized to other elderly populations.

Conclusions. We identified 38% prevalence of PA at a day-care facility in Japan. PA in this population was significantly associated with living alone and reduced protein intake and tended to be associated with depression. Given that nutritional intervention would generally be implemented after the onset of weight loss or undernutrition, assessment of appetite and related factors in the elderly should be of high priority to prevent undernutrition or malnutrition.

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CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

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Original

The Effect of Problem-Solving-Based Blood Glucose Management through Real-Time Self-Monitoring: A Case Study

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ABSTRACT *Background and Purpose* This study aimed to develop a nutrition education *program* for high school students and evaluate whether its implementation in regular classes improves health literacy (hereafter referred to as HL), health/eating knowledge, attitudes, and behaviors.*Methods* A non-randomized controlled trial was conducted with 98 high school students, divided into an intervention group (57 students) and a control group (41 students) at the class level. The primary endpoints were changes in HL and eating behaviors, while the secondary endpoints were changes in health awareness, attitudes, and knowledge. The intervention group participated in a total of four lessons using the nutrition education *program* composed of a card game, videos, three-color food cards, and cooking practice, while the control group received only a single lesson on health promotion. A pre- and post-intervention questionnaire surveys were conducted, along with a process evaluation based on feedback from both students and teachers. *Results* We conducted a per-protocol analysis on 47 students (83.9%) in the intervention group and 26 students (63.4%) in the control group who completed the follow-up. No significant changes in HL were observed during the intervention period in either group, and no significant differences were found between the groups. The significant improvements in the intervention group compared to the control group were observed only in the items related to health knowledge: the term BMI and its appropriate range, and lean body mass. In the process evaluation, high school students showed interest in the nutrition education program, and teachers expressed a desire to continue incorporating it into their classes. *Conclusion* No significant intervention effects were observed in HL, eating behaviors, or health awareness and attitudes. However, the process evaluation revealed positive feedback from students or teachers, indicating that the nutrition education *program* could be accepted and feasible in the school setting.

Keywords: high school students, health literacy, nutrition education *program*

INTRODUCTION

In recent years, health literacy (hereafter referred to as HL) has gained attention as a concept that enables individuals to obtain, understand, evaluate, and utilize health information (1), allowing them to maintain and promote their health throughout their lives (2). Developing HL through health education is not merely about understanding knowledge but is emphasized as a practical ability to recognize issues, make decisions, and take action using communication and social skills to maintain and improve health (3). On the other hand, during childhood and adolescence in which health risks have not still become apparent, it is presumed that active efforts to enhance HL are necessary from the perspective of acquiring healthy lifestyle habits. Research on HL has indicated a correlation between adolescent nutrition, physical activity, and HL, and intervention programs have been recommended to enhance HL (4). However, studies on HL (5,6) and observational research on HL among high school students (7,8) remain limited in Japan.

The high school curriculum guidelines (9) emphasize "thinking and decision-making skills" and state that students should "gather necessary information, acquire knowledge, and make judgments and decisions." Furthermore, they highlight the need for appropriate decision-making, behavioral choices, and the creation of environments that support health. High school represents the final stage in which school-based interventions can be implemented, while also being a period of increasing lifestyle diversity and prominent

nutritional challenges. Therefore, the development of a nutrition education *program* (hereafter referred to as *the program*) and intervention research are necessary.

Accordingly, this study reports on the development process and evaluation of *the program* aimed at improving HL, which can be incorporated into formal high school classes in Japan.

MATERIALS AND METHODS

2-1. Setting, Participants, and Ethical Considerations

A non-randomized controlled trial was conducted at two high schools, with 57 students assigned to the intervention group and 41 students to the control group at the class level.

We requested cooperation to the target schools, and after discussions and coordination with the school principals and teachers in charge, written explanations were provided to the students and their guardians to obtain informed consent. Participation was voluntary, and it was explained that responses would not affect grades or result in any disadvantages. Since the study was conducted during regular classes, to ensure anonymity during the consent process, documents were distributed and collected in sealed envelopes individually.

The study was reviewed and approved by the Research Ethics Committee of Aomori University of Health and Welfare (Approval No. 22059).

2-2. Study Procedure (Fig. 1)

The intervention and data collection period lasted four months, from November 2022 to March 2023. Pre- and post-intervention surveys were conducted to assess changes in HL, eating behaviors, health awareness and attitudes, and health knowledge.

The baseline survey was conducted for both groups before the first lesson, and the post-intervention survey was conducted at the end of the four-month *program* for the intervention group and at the same time for the control group. The primary endpoints were changes in HL and eating behaviors, while the secondary endpoints were changes in health awareness, attitudes, and knowledge. The intervention group participated in four lessons using *the program*, while the control group received a single lesson on health promotion. Additionally, a process evaluation was conducted based on feedback from students and teachers.

Date	Frequency	Time	Intervention †	Control ‡
2022 /11	1	50min	★Pre-survey Health promotion for adolescents(1) Card games(1) Videos	★Pre-survey Health promotion for adolescents
	2	50min	Health promotion for adolescents(2) Card games(2) Three-color food cards	
2023 /3	3	50min	Cooking practice	
	4	50min	Tasting and reflection ★Post-survey	★Post-survey

† The intervention group incorporated puberty health promotion, card games, videos, three-color food cards, and cooking practical sessions.
The lessons were structured as four 50-minute sessions, all conducted in person.
‡ The control group participated in one session of health promotion education for adolescence and completed a self-administered questionnaire survey with identification.

Fig 1. Research flow

2-3. Structure and Materials of the Nutrition Education Program (Fig. 2)

The program incorporated games, videos, three-color food cards, and cooking practice with the aim of fostering the ability to obtain, understand, evaluate, and utilize health information. To design the intervention content, we conducted a needs assessment and survey among high school students (10). The

survey (10) indicated that more than 50% of students were unaware of the "appropriate BMI range," yet many reported being "dissatisfied with their current weight." Furthermore, students identified "weighing themselves" and "cooking" as burdensome activities. In response, the lectures provided detailed explanations on appropriate BMI ranges and lean body mass, and students were guided through BMI calculations.

Based on the results, we developed *the program*. To maintain engagement, *the program* was not limited to lectures but also incorporated interactive elements such as games, videos, three-color food cards, and cooking practice to create a dynamic learning experience.

	Card games	Videos	Three-color food cards
Contents ・ Composition	Learn through games (health edition, knowledge edition, symptoms edition), become aware of your own and others' thoughts, physical discomfort, and bodily changes, and promote interactive communication.	University students created materials on the theme of "women's thinness," allowing high school students to deepen their understanding of the current state of thinness among Japanese women, the reasons for the increasing desire to be thin, DOHaD, and more, leading to greater awareness.	Understand food balance, learn its connection to cooking practice, and decide on ingredients for the cooking session. Create and use folded cards.

Fig 2. Content and structure of the nutrition education program

1) Card Game

Since positive feedback was received regarding the use of games in class, we determined that including the game in *the program* would not pose significant issues. The game's concept was to "imagine the feelings of others experiencing physical discomfort and communicate with them to foster mutual understanding."

High school students rarely have opportunities to discuss topics related to health or disease prevention. As a result, they struggle to verbalize symptoms of physical discomfort and remain dissatisfied with their weight despite lacking knowledge of appropriate BMI range. The game was designed to help students learn about "health" "knowledge " and "symptoms " . Through this interactive experience, students could recognize their own and others' perspectives, symptoms of discomfort, and bodily changes, while also improving their ability to communicate their symptoms to others (10).

2) Video

A lack of adequate health education has been identified as a factor contributing to underweight issues and insufficient energy intake among females (11). To address this, we requested university students to collaborate for creating a video on the theme of "female thinness." The video aimed to enhance understanding of the current state of underweight women in Japan and the reasons behind the growing desire to be thin, making the content easier to comprehend.

Many of the university students involved had previously experienced weight-related concerns and dieting during their high school years, recognizing the importance of effective initiatives to improve high school students' HL. Therefore, we incorporated a peer education approach, allowing university students to share important health-related information and promote a healthy body image among high school students. The goal was to create an engaging and informative learning experience.

The video consisted of two sections: a lecture and a cooking demonstration. Narration was provided by university students. The featured dish was "*Komatsuna* Fried Rice," which could be easily prepared using a single frying pan. The ingredients consisting of rice, *komatsuna* (Japanese mustard spinach), eggs, dried

whitebait, green onions, and sesame seeds, were selected to address deficiencies in energy, iron, and calcium intake.

3) Three-Color Food Cards

The three-color food card system which divides food into red, yellow, and green groups based on their functions is commonly used in school meal programs in primary and secondary schools. Therefore, we adopted it as a reference material because high school students are thought to be familiar with it.

In addition to learning about food balance, students were encouraged to apply this knowledge to cooking practice. *The program* was designed to help students understand food selection and meal planning, addressing difficulties they had previously identified, such as "cooking," "considering nutritional balance," and "planning meal combinations" (10).

The author proposed multiple (three to five) menu options, and students engaged in discussions to select a menu. They then determined the ingredients and portion sizes for their chosen meal.

4) Cooking Practice

The "Food and Nutrition" component of the home economics curriculum (12) emphasizes efficient cooking methods and their application to meal planning and menu creation. During discussions on the nutrition education *program*, teachers highlighted that students' dietary intake was often inadequate and that they had limited opportunities to cook. Based on this finding, we incorporated a cooking practice session where students selected their own menu.

The selected dish was "*Onigirazu*"—a sandwich-like rice ball that does not require shaping by hand. Using the knowledge gained from the three-color food card activity, students planned the ingredients and portions based on the following categories:

1. Yellow: Energy-providing foods
   - Rice, oil, mayonnaise
2. Red: Body-building foods (supporting blood, muscles, bones, and teeth)
   - Eggs, tuna, ham, seaweed, edamame
3. Green: Body-regulating foods (supporting skin, mucous membranes, and various body functions)
   - Spinach, tomatoes, cucumbers, carrots

For the cooking session, students were divided into groups of three per cooking station, with ingredients and utensils pre-prepared. The cooking process involved spreading half of the rice onto a sheet of seaweed, layering the chosen ingredients, adding the remaining rice, and folding the seaweed from all four corners. After explaining safety precautions, a demonstration was conducted, followed by hands-on cooking. Each student prepared their dish creatively and took photos of their completed meal.

2-4 Evaluation Methods

1) Questionnaire Composition

Basic information being collected only in the pre-intervention survey included grade level, age, gender, height, weight, and club activity participation. The pre- and post-intervention surveys assessed HL, eating behaviors, health awareness and attitudes, health knowledge, daily routines, exercise frequency, and menstrual records. HL was measured using the Communicative and Critical Health Literacy (CCHL) scale (13).

2) Data Collection and Analysis

The questionnaire was administered twice before and after the intervention in a self-reported format. Survey forms were distributed and collected in sealed envelopes to ensure confidentiality. Participants who were absent, left questions blank, or failed to provide their names were excluded from the analysis. A per-protocol analysis was conducted for the remaining participants.

Intra-group comparisons and inter-group comparisons were analyzed for HL, eating behaviors, health awareness and attitudes, health knowledge, daily routines, exercise frequency, and menstrual records.

Statistical power was set at 80%, with a 5% alpha error. A medium effect size (d = 0.50) was used to calculate a required sample size of 26 participants per group using G*Power 3.1.9.7.

For group comparisons of HL scores and baseline characteristics, t-tests were conducted. Pre- and post-intervention comparisons within groups were analyzed using the Wilcoxon signed-rank test for ordinal variables and McNemar's test for nominal variables. Mann-Whitney U tests were used for inter-group comparisons of changes. Missing data were treated as missing values. Statistical analyses were performed using IBM SPSS Statistics Ver. 29® (IBM Japan), with a significance level of <5% (two-tailed).

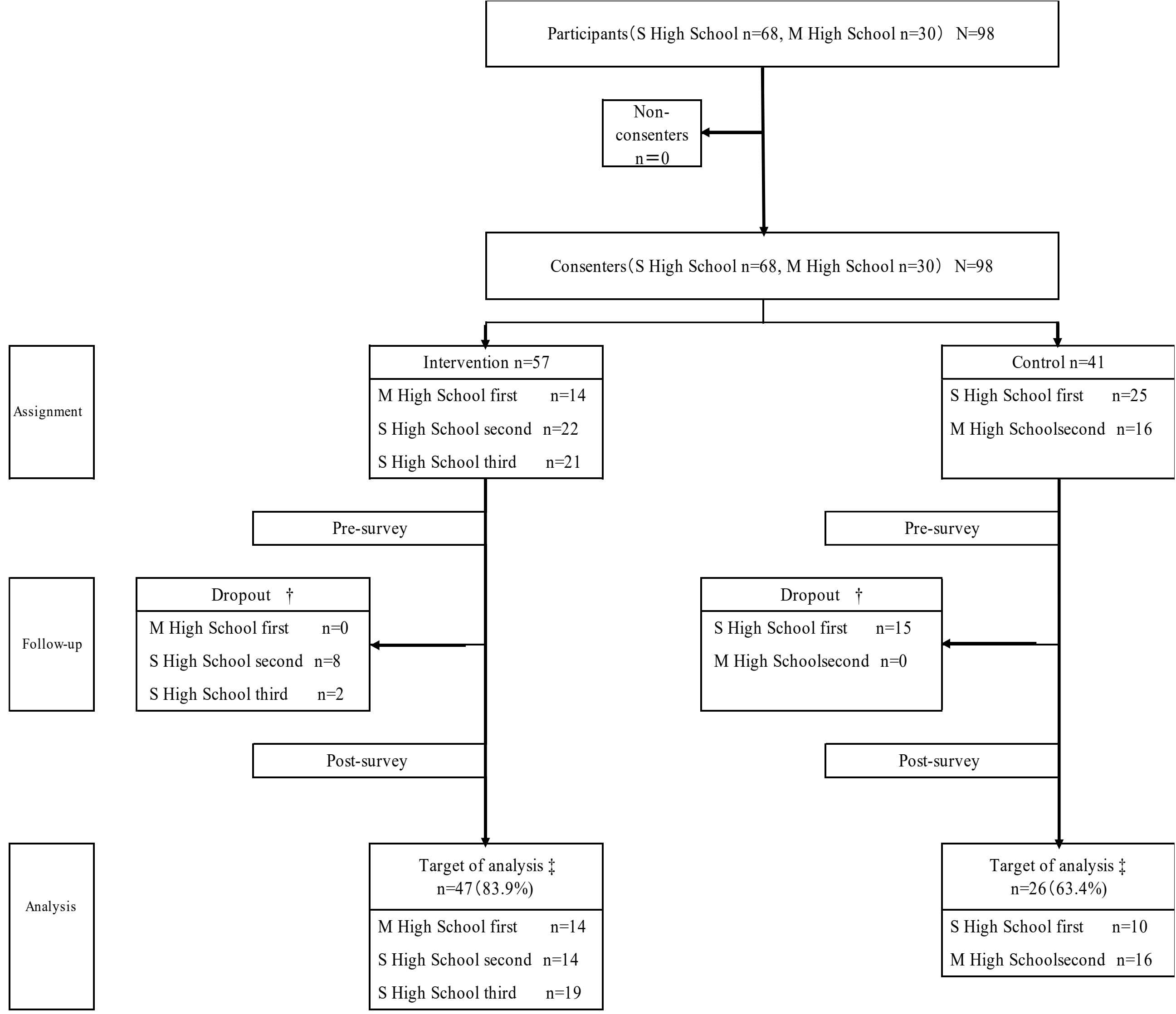
3) Process Evaluation

Process evaluation was conducted based on *program* implementation status, student feedback, and teacher comments. Free-response feedback from students was categorized into themes related to games, lessons, and cooking practice. Teacher feedback was classified into observations of student engagement and opinions on

the program.

RESULTS

The number of participants included in the analysis was 47 (83.9%) in the intervention group and 26 (63.4%) in the control group (Fig. 3).



It was targeted at first, second, and third-year students from five classes at S High School and M High School.
Consent was obtained from all participants, and the allocation was done by high school and grade (by class)
The dropout rate was 28% in the intervention group and 60% in the control group at S High School, while at M High School, the dropout rate was 0% in both the intervention and control groups.
† Dropout refers to those who were absent from the classes, attended but did not fill out the questionnaire items, or filled out the questionnaire but did not provide their name, making it impossible to conduct pre- and post-comparisons.
‡ Analysis subjects refer to those who are identified by name and have no missing responses.

Fig 3. Comparison test flow chart

3-1 Basic Information of Participants

No significant differences were observed between the intervention and control groups (Table 1).

Table 1. Basic attributes

	Intervention		Control	
	n=47	SD	n=26	SD
Age	16.6	0.79	16.4	0.80
Height（cm）	163.5	9.16	160.0	8.07
Weight（kg）	57.0	12.66	54.3	13.99
Body Mass Index（kg/m²）	21.3	4.12	21.0	3.76
Gende Boys	20人(43%)		5人(24%)	
Girls	27人(57%)		21人(76%)	
Sports Club	21人(45%)		11人(42%)	
Non-Sports Club	26人(55%)		15人(58%)	

N=73

Age, height, weight, and BMI are presented as the mean and standard deviation.

Gender and club activity are presented as the number of participants (%).

3-2 Primary Endpoint

1) HL Score (Table 2, 3)

The mean HL score before the intervention was 3.85 (SD 0.86) in the intervention group and 4.01 (SD 0.47) in the control group, with no significant differences between the groups at baseline.
After the intervention, the mean HL score was 3.94 (SD 0.62) in the intervention group and 3.90 (SD 0.58) in the control groups.

There were no significant changes in the mean HL scores before and after the intervention within each group, nor were there significant differences between the intervention and control groups.
A comparison of pre-intervention HL score averages by grade level showed that first-year students had a mean of 3.91 (SD 0.62) and second-year students had a mean of 3.79 (SD 0.68), with no significant differences between them.

Table 2. Comparison of overall distribution of health literacy scores

		Total HL score														
		5-9		10-14		15-19		20-24		25		Median	Interquartile range	$P^{\dagger}$	$P^{\ddagger}$	$P^{\S}$
		n	(%)	n	(%)	n	(%)	n	(%)	n	(%)					
Whole (n=73)	Pre	2	(2.7)	2	(2.7)	29	(39.7)	34	(46.6)	6	(8.2)	20	(17 - 22)	0.728	-	-
	Post	1	(1.4)	4	(5.5)	26	(35.6)	35	(47.9)	7	(9.6)	20	(18 - 21)			
Intervention (n=47)	Pre	2	(4.3)	2	(4.3)	18	(38.3)	20	(42.6)	5	(10.6)	20	(17 - 22)	0.862	0.862	0.839
	Post	1	(2.1)	2	(4.3)	17	(36.2)	21	(44.7)	6	(12.8)	20	(18 - 21)			
Control (n=26)	Pre	0	(0.0)	0	(0.0)	12	(46.2)	14	(53.8)	0	(0.0)	20	(18 - 22)	0.404	-	-
	Post	0	(0.0)	2	(7.7)	8	(30.8)	15	(57.7)	1	(3.8)	20	(18 - 22)			

†:For the comparison between pre-intervention and post-intervention, the Wilcoxon signed-rank test was conducted.

‡:For the comparison between the intervention group and the control group before the intervention, the Mann-Whitney U test was conducted.

§:For the comparison between the intervention group and the control group after the intervention, the Mann-Whitney U test was conducted.

Table 3. Comparison of mean health literacy scores

		n	HL Mean	SD	*P*
H L Before and after	Intervention	47	0.094	0.62	0.11 †
(Post - pre)	Control	26	-0.108	0.43	
Intervention	Post	47	3.94	0.74	0.31 ‡
	Pre	47	3.85	0.86	
Control	Post	26	3.90	0.58	0.22 ‡
	Pre	26	4.01	0.47	
Pre Boys		25	3.95	0.83	0.47 †
Pre Girls		48	3.90	0.74	
Pre first-year students		24	3.91	0.62	0.68 †
Pre second-year students		30	3.79	0.68	
Pre third-year students		19	3.93	0.85	

N=73

† For the between-group comparison, the Mann-Whitney U test was conducted.

‡ For the within-group comparison, the Wilcoxon signed-rank test was conducted.

2) Eating Behavior, Daily Routine, Exercise Frequency, and Menstrual Records(Table.4)

There were no significant changes before and after the intervention within either the intervention or control groups, and no significant differences were observed between the groups.

Table 4. Food behavior, lifestyle, exercise frequency, and menstrual records

		Intervention（n=43）			Control（n=26）			Intervention vs Control	
		Pre	Post		Pre	Post		Pre	Post
		n %	n %	$P^{\dagger}$	n %	n %	$P^{\dagger}$	$P^{\ddagger}$	$P^{\ddagger}$
Breakfast	Eat almost every day	34 (72.4)	33 (70.3)	0.317	23 (88.5)	21 (80.8)	0.48	0.118	0.293
	Eat 2-3 times a week	9 (19.1)	9 (19.1)		2 (7.7)	4 (15.4)			
	Hardly eat	4 (8.5)	5 (10.6)		1 (3.8)	1 (3.8)			
Lunch	Eat almost every day	42 (89.3)	42 (89.3)	0.317	25 (96.2)	26 (100.0)	0.317	0.298	0.087
	Eat 2-3 times a week	2 (4.3)	3 (6.4)		1 (3.8)	0 (0.0)			
	Hardly eat	3 (6.4)	2 (4.3)		0 (0.0)	0 (0.0)			
Dinner	Eat almost every day	45 (95.7)	46 (97.9)	0.564	25 (96.2)	23 (88.5)	0.157	0.933	0.093
	Eat 2-3 times a week	2 (4.3)	1 (2.1)		1 (3.8)	3 (11.5)			
	Hardly eat	0 (0.0)	0 (0.0)		0 (0.0)	0 (0.0)			
Cooking	Almost every day	5 (10.6)	7 (14.9)	1	4 (15.4)	2 (7.7)	0.014	0.723	0.361
	Make it 2-3 times a week	23 (48.9)	19 (40.4)		12 (46.2)	10 (38.5)			
	Hardly cook	19 (40.5)	21 (44.7)		10 (38.5)	14 (53.8)			
Bedtime (weekdays)	Before 10p.m	4 (8.5)	3 (6.4)	0.74	5 (19.2)	2 (7.7)	0.059	0.245	0.518
	10 to 11p.m	6 (12.8)	8 (17.0)		4 (15.4)	6 (23.1)			
	11p.m to midnight	21 (44.7)	19 (40.4)		10 (38.5)	10 (38.4)			
	After midnight	16 (34.0)	17 (36.2)		7 (26.9)	8 (30.8)			
Bedtime (holidays)	Before 10p.m	3 (6.4)	3 (6.4)	1	3 (11.5)	3 (11.5)	0.705	0.604	0.697
	10 to 11p.m	4 (8.5)	4 (8.5)		3 (11.5)	2 (7.7)			
	11pm - midnight	17 (36.2)	17 (36.2)		8 (30.8)	9 (34.6)			
	After midnight	23 (48.9)	23 (48.9)		12 (46.2)	12 (46.2)			
Wake-up time (weekdays	Before 6 o'clock	19 (40.4)	15 (31.9)	0.034	8 (30.8)	10 (38.5)	0.705	0.849	0.519
	From 6 to 7	20 (42.6)	24 (51.0)		16 (61.5)	13 (50.0)			
	From 7 to 8	8 (17.0)	6 (12.8)		2 (7.7)	1 (3.8)			
	After 8 o'clock	0 (0.0)	2 (4.3)		0 (0.0)	2 (7.7)			
Wake-up time (holidays)	Before 6 o'clock	2 (4.3)	1 (2.1)	0.331	3 (11.5)	3 (11.5)	0.763	0.922	0.877
	From 6 to 7	10 (21.3)	8 (17.0)		2 (7.7)	3 (11.5)			
	From 7 to 8	15 (31.9)	18 (38.3)		10 (38.5)	7 (26.9)			
	After 8 o'clock	20 (42.5)	20 (42.6)		11 (42.3)	13 (50.1)			
Sleep time (weekdays)	Less than 6 hours	19 (40.4)	21 (44.7)	0.763	9 (34.7)	7 (26.9)	0.763	0.115	0.043
	6 to 7 hours	21 (44.7)	18 (38.3)		7 (26.9)	10 (38.5)			
	7 to 8 hours	6 (12.8)	7 (14.9)		5 (19.2)	4 (15.4)			
	Over 8 hours	1 (2.1)	1 (2.1)		5 (19.2)	5 (19.2)			
Sleep time (holidays)	Less than 6 hours	3 (6.4)	4 (8.5)	0.334	1 (3.8)	2 (7.7)	0.386	0.232	0.323
	6 to 7 hours	13 (27.7)	16 (34.1)		5 (19.2)	5 (19.2)			
	7 to 8 hours	17 (36.1)	12 (25.5)		9 (34.6)	9 (34.6)			
	Over 8 hours	14 (29.8)	15 (31.9)		11 (42.4)	10 (38.5)			
Exercises other than physical education	Hardly ever	16 (34.0)	17 (36.2)	0.763	4 (15.4)	4 (15.4)	0.655	0.065	0.035
	2-3 days a week	16 (34.0)	15 (31.9)		9 (34.6)	8 (30.8)			
	Every day	15 (32.0)	15 (31.9)		13 (50.0)	14 (53.8)			
Menstruation	Recording, aware of the cycle	12 (44.4)	9 (33.3)	0.681	8 (38.1)	9 (42.9)	0.705	0.894	0.467
	Recording, but not aware of the cycle	2 (7.4)	4 (14.8)		4 (19.0)	4 (19.0)			
	Not recording, aware of the cycle	10 (37.0)	12 (44.5)		8 (38.1)	6 (28.6)			
	Not recording, and not aware of the cycle	3 (11.1)	2 (7.4)		1 (4.8)	2 (9.5)			

N=73

Menstrual record =48

†: A comparison of pre- and post-intervention within the group was conducted using the Wilcoxon signed-rank test.

‡: A comparison of pre- and post-intervention between groups was conducted using the Mann-Whitney U test.

3-3 Secondary Endpoint

1) Health Awareness and Attitudes (Table 5)

No significant changes were observed before and after the intervention within either the intervention or control groups, nor were there any significant differences between the groups.

Table 5. Health awareness

		Intervention (n=47)			Control (n=26)			Intervention vs Control	
		Pre	Post		Pre	Post		Pre	Post
		n %	n %	$P^{\dagger}$	n %	n %	$P^{\dagger}$	$P^{\ddagger}$	$P^{\ddagger}$
My weight	Not satisfied	34 (72.3)	30 (63.8)	0.102	19 (73.1)	16 (61.5)	0.083	0.947	0.847
	satisfied	13 (27.7)	17 (36.2)		7 (26.9)	10 (38.5)			
My condition	Not satisfied	26 (55.3)	16 (34.0)	0.008	10 (38.5)	7 (26.9)	0.102	0.29	0.533
	satisfied	21 (44.7)	31 (66.0)		16 (61.5)	19 (73.1)			

N=73

†: A comparison within the group was conducted using the McNemar test.

‡: A comparison between groups was conducted using the chi-square test.

In the intervention group, approximately 90% of participants rated five out of seven items—"adequate sleep," "balanced physique," "moderate exercise," "eating three meals a day," and "paying attention to dietary balance", as important (either "very important" or "important") both before and after the intervention.

2) Health Knowledge (Table 6)

In the intervention group, significant changes were observed before and after the intervention regarding knowledge of "the term BMI," "the appropriate BMI range," "lean body mass," and "one's own physical condition."

When comparing between groups, significant differences were found in "The term BMI", "The appropriate BMI range", and "Lean body mass".

Table 6. Health knowledge

		Intervention (n=47)			Control (n=26)			Intervention vs Control	
		Pre	Post		Pre	Post		Pre	Post
		n %	n %	$P^{\dagger}$	n %	n %	$P^{\dagger}$	$P^{\ddagger}$	$P^{\ddagger}$
The term BMI	Don't know	14 (29.8)	3 (6.4)	<0.001	8 (30.8)	8 (30.8)	1	0.931	0.006
	Iknow	33 (70.2)	44 (93.6)		18 (69.2)	18 (69.2)			
Normal BMI range	Don't know	34 (72.3)	13 (27.7)	<.001	20 (76.9)	17 (65.4)	0.083	0.671	0.002
	know	13 (27.7)	34 (72.3)		6 (23.1)	9 (34.6)			
Lean body mass	Don't know	40 (85.1)	23 (48.9)	<.001	24 (92.3)	19 (73.1)	0.025	0.373	0.047
	know	7 (14.9)	24 (51.1)		2 (7.7)	7 (26.9)			

N=73

†: A comparison within the group was conducted using the McNemar test.

‡: A comparison between groups was conducted using the chi-square test.

3-4 Process Evaluation

1) Students' Free Responses

The following comments were extracted as free responses.

(1) Card Game

- There were many words I didn't know.
- Using the symptom cards in the game helped me realize what symptoms I might have.
- If I know, I can approach things confidently and positively.
- I learned not to be swayed by incorrect information and to gather reliable information.

(2) Lessons

- I didn't know the BMI criteria before.
- If you focus too much on weight, it can lead to amenorrhea or osteoporosis.
- There is a lot of information, and I need to decide whether it's true or false.

(3) Cooking Class

- I feel like I could make this at home.
- I used to skip breakfast, but I realized that eating breakfast makes my day more enjoyable.
- I found cooking fun.
- Since I love eating, I felt very happy during the cooking class.
- I will try to eat more and increase my portion sizes.
- I plan to cook at home, even if it's just a little.
- I will start by being more conscious of eating breakfast and small things.
- I think I will try cooking at home.

2) Teacher Feedback

The following comments were extracted as free responses.

(1) Student Behavior

- In cooking classes, students showed creativity and initiative, and they became more interested in food.
- After the cooking class, some students brought their own lunch or snacks, saying, "I can make this with what I have at home."
- Students felt a sense of familiarity with the video created by college students and showed interest in the simple recipes.

(2) Nutrition Education Program

- *The program* catered to students who had difficulty engaging or had varying interests, and continued participation led to knowledge retention. I want to incorporate it into future lessons.
- Before participating in *the program*, students only used the height and weight scales in the hallway of the health room. After the program, they were seen calculating their BMI after measuring their height and weight.

DISCUSSION

4-1 Health Literacy (HL)

1) Comparison with Previous Research Using the Measurement Scale

In this study, the pre-intervention mean HL score for the intervention group was 3.85 (SD 0.86), and for the control group, it was 4.01 (SD 0.47). This is comparable to the study by Kasahara et al. (8), which reported average HL scores of 3.66 (SD 0.81) for males and 3.71 (SD 0.68) for females. There was no significant difference when comparing the results of this study with those of Kasahara et al.

2) Within-group and Between-group Comparisons

For each item, there were no changes observed within the intervention group or the control group when comparing pre- and post-intervention surveys. Additionally, no significant differences were found when comparing the pre- and post-intervention surveys between the intervention and control groups. Regarding the interactive HL measurement scale, 34.0% of respondents agreed somewhat, and 55.3% strongly agreed with the information gathering items. However, there was significant variability in responses for information extraction, communication, critical HL, reliability judgment, and decision-making.

It has been reported that previous school health education primarily focused on the transmission of basic health knowledge, without providing skills to engage in community activities (14). *The program* developed in this study aimed to go beyond knowledge transmission, incorporating activities designed to foster active engagement and dialogue, allowing students to participate interactively.

Despite the positive intent and structured design of *the program*, the absence of significant changes in HL, eating behavior, and attitudes towards health suggests the need for further exploration and refinement of such *programs*. The lack of significant effects may be related to factors such as the duration of the intervention, the specific content delivered, or the engagement level of students with the materials and activities provided.

4-2 Eating Behavior and Eating Habits

1)Teacher Feedback

Teachers reported positive outcomes, such as students becoming more interested in cooking and bringing breakfast or lunch from home. In particular, students showed creativity during cooking classes, taking an active interest in food preparation. Feedback highlighted that *the program* helped students gain interest in meal planning and cooking, making it an important foundation for maintaining healthy eating habits.

3) Cooking Frequency and Future Implications

Around half of the students responded that they cooked "2-3 times a week," which may be influenced by the busy schedules of high school students with academics and extracurricular activities. However, it is expected that after graduation, when students often live independently, the frequency of cooking will increase. The hands-on cooking experiences and food-related activities in *the program* help reduce the burden and anxiety around cooking, encouraging students to enjoy the process. Feedback included comments like "cooking is fun" and "it's delicious when you eat together," which suggests that *the program* fostered a positive attitude toward cooking and shared meals.

4-3 Health Awareness and Attitudes

1)Pre- and Post-Intervention Health Awareness

Around 90% of students in both the intervention and control groups indicated that aspects such as "sleep" and "balanced body composition" were important both before and after the intervention. This suggests that a baseline awareness already existed among students. The program's interactive nature, which encouraged listening to others' opinions and engaging in two-way communication, was thought to reinforce these attitudes and maintained health awareness.

2)Body Weight and Satisfaction

Following the intervention, more students expressed satisfaction with their body weight, and learning how to calculate BMI and understand the criteria helped correct misconceptions about body image and weight. Additionally, increased satisfaction with their physical condition was observed, as students were able to reflect on and understand their symptoms and health status through discussions with peers.

4-4 Health Knowledge

1)Knowledge Acquisition and Behavior Change

A positive change in health knowledge was observed in the intervention group, especially concerning decision-making processes related to health. This suggests a direct effect of the program on students' knowledge. Acquiring knowledge about health is crucial for the prevention of lifestyle diseases. The process of actively engaging in discussions and learning from each other is expected to strengthen problem-solving abilities, and repeated learning is essential for effective retention.

2) Comparison with Other Studies

The LifeLab program based at Southampton University Hospital has reported lasting changes in HL and critical judgment related to personal behaviors (16). This program, combined with other studies, has led to an increase in awareness and positive changes in attitudes regarding adolescent health. Although HL improved, knowledge alone did not result in behavioral changes, suggesting that sustainable behavior change requires further research. Teachers also need training in supporting students to make healthier choices.

3)Further Considerations

While knowledge about health improved through the program, there were no substantial behavioral changes or lasting transformation in health habits. This underscores the challenge of achieving long-term behavior change. For sustainable change, students need to develop the ability to identify challenges in their own lifestyles and to select and apply accurate health information. It is also important to consider integrating family support and feedback into the program. Health education that focuses on information collection, discussion, critical thinking, and decision-making can play a significant role in fostering long-term health literacy.

4-5 Process Evaluation

In this study, no negative feedback was received about *the program*. On the contrary, the majority of the participants found *the program* to be enjoyable and engaging, providing a fun and interactive learning experience. Instead of passively acquiring knowledge, the students were able to engage in active and dialogic learning, creating a relaxed atmosphere where they could freely discuss health issues and physical conditions. This type of two-way communication was useful in drawing out the students' interest and attention.

1) Students' Free Descriptions

(1) Card Game

Students reported that the card game helped them recognize their own symptoms and approach health issues with a positive attitude. The game made them aware that symptoms can vary from person to person, which led to an appreciation for different perspectives. The card game allowed students to gain a deeper understanding of their symptoms, and they recognized the importance of gathering accurate information for health improvement.

(2) Class

During the lessons, students were able to select actions and plans to improve their health based on the information provided, which helped them critically evaluate and choose reliable information. The use of active learning in high school classes is expected to led students to think more comprehensively, expand their ideas, and deepen their understanding. Dialogic learning seemed particularly effective, and students were able to acquire information, evaluate it, make decisions, and apply critical thinking.

(3) Cooking Classes

In cooking classes, even students who usually participated less in regular lessons became more active and engaged. The cooking activities allowed students to set realistic and achievable health goals. Moreover, they learned the joy of cooking and the importance of eating meals. Recent trends, such as the rising popularity of "*Onigirazu"* (rice ball wraps), which is easy to prepare and eat, fit well into the curriculum and helped reduce skipping meals and provide a convenient snack option. These cooking lessons seemed as valuable in promoting health.

2) Teacher Feedback

Teachers noted that students' interest and engagement in the program increased as they learned about their own bodies. Given that high school graduation marks a time when students' lifestyle habits change significantly, introducing such programs is expected to enhance students' motivation for health improvement. Teachers expressed positive feedback, and some mentioned they desire to keep using *the program* in the future. They also felt that *the program* could be easily implemented in high school classes.

4-6 Limitations of the Study

To assess the true effects of the intervention, a randomized controlled trial would be ideal. However, conducting such an experimental design in real-world school settings is often challenging. Therefore, this study adopted a quasi-experimental design with a non-randomized control group, conducting the intervention in two high schools with practical field constraints. The participants in this study may have had a high level of interest or concern about food and nutrition, which could have influenced the outcomes. While the program was designed to encourage active participation and knowledge transfer, there was insufficient consideration of activities to foster critical thinking during the application of this knowledge. Further research could improve these aspects to strengthen t*he program's* effectiveness.

CONCLUSION

This study developed a nutrition education *program* aimed at improving health literacy (HL) among high school students and implemented it through an intervention trial in classrooms. However, it was not possible to demonstrate whether *the program* led to healthier eating habits. On the other hand, students were able to actively and dialogically engage in learning, gain a better understanding of their own health, and develop an interest in the nutrition education *program*. Teachers expressed interest in continuously incorporating *the program* into their lessons, and the feasibility of implementation in schools was favorable.

For high school students, it was crucial to cultivate the ability to obtain, understand, evaluate, and apply health information, as well as to develop the process of critical thinking to address health issues related to their lifestyles. Introducing health education in the classroom and allowing students to acquire knowledge was effective and highly satisfactory. It is believed that this would lead to improved HL and support long-term, healthy behaviors and appropriate decision-making in the future.

In light of this, to enhance the feasibility of introducing the developed nutrition education *program* in classrooms, standardization of t*he program's* procedures and content is necessary. *The program* will be refined to improve HL from the perspective of developing problem-solving skills for health issues. A more sophisticated research design will be used to conduct a follow-up intervention study.

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Original

Cooking-integrated Nutrition Education Program Affects Stages of Behavior Change among Fourth-Grade Students in Vegetable, Fruit and Sugar-Sweetened Beverage Consumption

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ABSTRACT *Background.* Studies have shown that integrating cooking activities with nutrition education can improve children's preferences for and intake of fruits and vegetables (FV). However, evidence regarding its impact on sugar-sweetened beverages (SSB) and stages of behavior change remains limited. This pilot study aimed to evaluate the effect of an experiential cooking and nutrition education intervention on stages of behavior change related to FV and SSB consumption among elementary school students. *Methods.* A total of 35 fourth-grade subjects were recruited through purposive sampling. Using crossover design, the effects of cooking integrated nutrition education curriculum on nutrition knowledge (NK), selecting attitude (AT), and consuming behavior (BE) towards FV and SSB, as well as stages of behavior change (SOC), were examined. Subjects were divided into Group A (n=16) and Group B (n=19), randomized to begin with either cooking or nutrition education curriculum, before switching to the alternate curriculum. The intervention comprised eight lessons over two 4-week periods, with five assessments conducted at pretest (W_0), midterm test (W_4), posttest (W_8), follow-up test (W_{10}) and a final test (W_{20}). *Results.* Results indicated significant improvements in NK and BE score (p<.05). The overall intervention facilitates subjects' behavior change to progress to a later stage by 20%, 25.7% and 37.1% towards vegetables, fruits and sugar sweetened beverages, respectively. *Discussion.* The intervention improved NK with retention and carryover effects. BE increased especially when nutrition education preceded cooking. Cooking was more effective for FV, while education reduced SSB. *Conclusion.* The integration of cooking activities with nutrition education effectively supported students' advancement through stages of behavior change and yielding stronger outcomes, particularly in reducing unhealthy SSB consumption. This approach shows promises for promoting healthy eating behaviors in elementary school settings.

Keywords: Cooking program, Knowledge Attitude Behavior (KAB), Stages of Change, Fruits and Vegetables, Sugar-sweetened Beverages

INTRODUCTION

The epidemic of childhood obesity has emerged as an important public health problem worldwide. In 2016, the prevalence of childhood obesity increased at an alarming rate. This is associated with various diseases such as type 2 diabetes mellitus, hypertension, nonalcoholic fatty liver disease, Obstructive sleep apnea, and dyslipidemia later in their adult age (1).

In Taiwan, the prevalence rate of overweight and obesity among elementary school children aged 6-12 years old was increasing substantially since year 2008 (25.0%) to 2013 (30.4%) but decreasing steadily to 27.6% in 2017 (2). Despite the decreasing trend being observed over the past five years, Taiwan Nutrition and Health Survey in Taiwan Elementary School Children (NAHSIT IV Children) in year 2012 reported that the highest prevalence rate of obesity was noticed among 4th grade male students (17.1%) and 5th grade female students (15.6%) (3).

Diet plays an important role in preventing overweight and obesity. Study reviewed that there was an inverse association between a prudent/healthy dietary pattern and overweight/obesity risk and a positive association between a western/unhealthy dietary pattern and overweight/ obesity risk (4). Fruits and

vegetables (5), fiber (6), fat (7), fast food (8) and sugary drinks (9) are included but not limited to as important dietary determinants of obesity in childhood and adolescence. It is plausible that dietary pattern that are energy- and fat- dense but low in fiber may be predictive of later overweight and obesity in children (10). By consuming recommended amounts of fruits, vegetables, and whole grains would result in a healthier dietary pattern. Unfortunately, children often have poor compliance with dietary guidelines for these foods (11). Furthermore, evidence that decreasing sugar- sweetened beverage (SSB) consumption will reduce the prevalence of obesity and obesity- related diseases is increasingly clear (9).

Health Promotion Administration, Ministry of Health and Welfare of Taiwan has implemented the daily dietary guidelines to combat the problem of imbalance diet among school-aged children. In term of consumption of fruits and vegetables, dietary patterns of students reflected that 72.9% of students do not fulfill the recommended intake which is five servings per day. Among all, only 12.3% of school-aged children fulfill the daily recommended intake of three servings of vegetables and two servings of fruits at the same time. Nutrition and Health Survey in Taiwan Elementary School Children (NAHSIT II Children) year 2001-2002 documented that energy intake from carbohydrate was inadequate. In addition, 7.2% of the carbohydrate source was from sugar sweetened beverages. This issue should be addressed wisely because the more sugar- sweetened beverage (SSB) consumed, both weight and body mass index (BMI) of children will increase (12,13).

Many studies focused on the evaluation of nutrition education intervention towards students' knowledge, attitude and behavior, most studies showed the same outcome that there was a significant increase in students' knowledge, attitude and behavior. The concept of experiential learning such as cooking, gardening and tasting has emerged and incorporating in nutrition education program which helps to improve the preference of students towards fruits and vegetables (14,15,16).

Unfortunately, there are lack of researches done in assessing stages of behavioral change after implementing cooking-integrated nutrition education intervention. It is utmost important as stage-tailored intervention concept is believed to increase the effectiveness of nutrition education program (17). Thus, this study aims to evaluate the effectiveness of experiential cooking program and nutrition education intervention through nutrition knowledge (NK), selecting attitude (AT) and consuming behavior (BE). Also, its effect towards stages of behavioral change (SOC) on consumption of fruits, vegetables and sugar sweetened beverages were evaluated as well.

MATERIALS AND METHODS

Study Design This was a 5-months evaluation of experiential cooking and nutrition education program. By adapting cross-over design concept in nutrition education intervention (18), the research design called for a random assignment of all 41 subjects to either Group A (n=20) or Group B (n=21). A total of 8 lessons of curriculum were delivered in two 4-week periods. Group A subjects were randomized to begin with cooking curriculum in first 4-weeks period, whereas Group B begin with nutrition education curriculum, and then both groups switch to the remaining curriculum in second 4-weeks period. Both programs had a similar delivery and length. Each subject completed a pretest (W_0), a midterm test (W_4), a posttest (W_8), a follow-up test (W_{10}) and a final test after semester break (W_{20}) as shown in Figure 1.

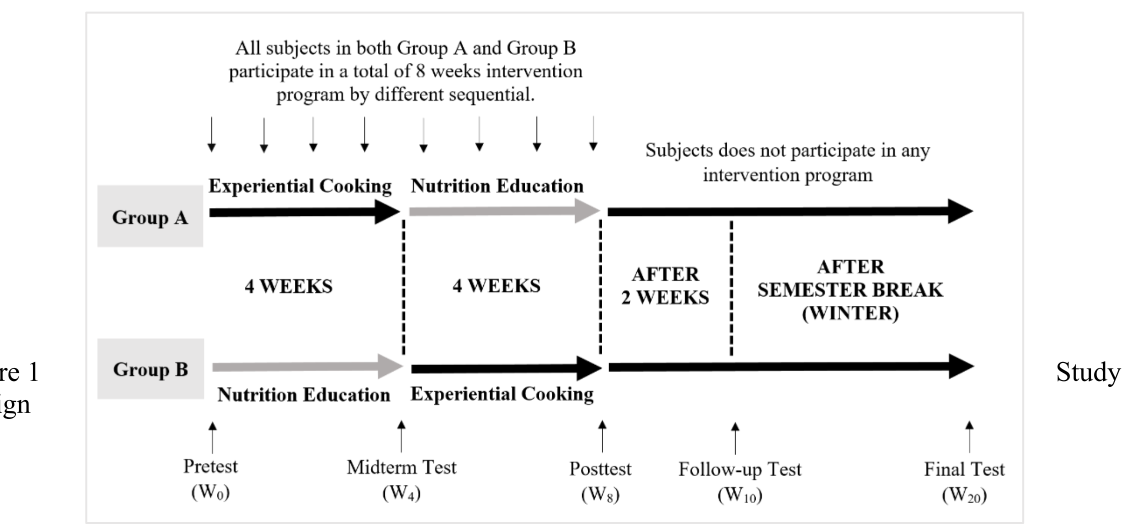

Figure 1 Study design

Participants and Recruitment The evaluation included 41 subjects selected through purposive sampling

from an elementary school located in *Daliao* district, Kaohsiung. Application was opened for all Fourth-grade students one month prior to the intervention. Students applied to participate according to their own willingness. The inclusion criteria for the program were subjects without food allergy history especially milk, eggs and nuts. Subjects with asthma history were also excluded. Application packets, which contain informed consents for both parents and subjects were distributed to selected subjects.

Ethical Approval This research was reviewed and approved by the institutional review board in Antai Medical Care Corporation Antai Tian-Sheng Memorial Hospital. (Protocol No. 18-109-B)

Interventions Both experiential cooking and nutrition education curriculum were discussed and designed by nutrition teacher and third year nutrition students, incorporating the service- learning courses that is selective among undergraduate students. The experiential cooking curriculum was designed to focus primarily on simple hands on preparing hand- hold food rather than cooking skills. However, simple cutting and preparing skills were also imparted to subjects to ensure that they could experience food ingredients. The nutrition education curriculum was designed with aims of instilling nutrition knowledge towards subjects through different teaching methods including didactic instruction, drama in education, practice instruction and team-games-tournament. Both curriculums were reviewed by registered dietitian to ensure that recipes created were aligned with principles of good nutrition and the appropriateness of nutrition knowledge imparted. Lessons were led by nutrition students and they were required to went through food tasting and rehearsal training at least 2 times (1 hour per time) by nutrition teacher and registered dietitian. The whole program consisted of 8 30-minutes lessons taught during the self-study period in the morning. All lessons were taught in Mandarin and nutrition principles illustrated was aligned with new version of Dietary Guideline of Taiwan in 2018. Subjects were formed into teams of four to five, which were maintained throughout the course, to practice food preparing skills, and carry out team games throughout the lessons.

Instruments and Measures The evaluation of the intervention was assessed by self-developed survey questions. The survey was first developed by adapting questions from publicly available sources (19,20), then was reviewed by five experts in nutrition education and/or public health to ensure content validity and audience appropriateness. A total of 50-items including subject's demographic data (8 items), NK (15 items), AT (12 items), BE towards FV and SSB (12 items) and SOC (3 items) were collected through the written surveys. The survey was pilot tested among 65 subjects which were not participated in the study. Average item discrimination index (D) for nutrition knowledge-testing questions reported was 0.41, considered very usable (21). Internal consistency was assessed with Cronbach alpha coefficient; scale structure was analyzed using principle components extraction with varimax rotation. Cronbach alpha coefficient of 0.72 and 0.84 were achieved for both scale of selecting attitude and consuming behavior which were measured in five-point Likert scale. Due to the poor time frame estimation among elementary school students, excluding question regarding time frame in stage of behavior change was necessary. Thus, instead of five stages, three stages of behavior change were used including precontemplation (PC, do not know/plan to change), contemplation/preparation (C/P, plan to change) and action/maintenance (A/M, have changed unhealthy behavior) (3).

Survey Administration Surveys were administered for all subjects present on the day of administration. Teacher was present in the classroom but not directly involved in survey administration. Instructions for all parts of survey were read aloud by researchers in preferred language (Mandarin) with guidance to students to complete the rest of the page independently. Measuring instrument (10cm circumference bowl) was used to guide students on the portions of fruits and vegetables being consumed. Completion of survey took around 20 minutes. Upon collecting the survey form, researcher ensure that all parts were answered.

Data Analysis Subjects who missed out at least two lessons or surveys that contains one third uncomplete parts will be excluded for analysis. Statistical analysis was performed using IBM SPSS Statistics Version 22 (SPSS Inc., Chicago, IL, USA). For each scale except for the SOC, item responses were summed to create a scale score. Desired outcomes were noted by higher scores. Wilcoxon Signed Rank test was used to compare the median difference of NK, AT, and BE from pretest to final test within both Group A and B. Median difference between Group A and B were tested using Mann-Whitney U test. Subjects' behavioral stage distribution at each time point and the congruence of stages between pretest, midterm test and posttest were examined using frequency distribution and cross-tabulations. $P<.05$ were considered statistically significant.

RESULTS

Demographics Of all 41 applicants, 35 subjects remained and completed the whole program and surveys, survey response rate was 85.3%. Subjects were predominantly female (68.6%), with cooking experience (71.4%) and cooking interest (97.1%) (Table 1). Data between Group A and B showed no statistically different ($p>.05$), which are comparable. Subjects' stage of behavioral change for vegetables and fruits were predominantly at precontemplation stage whereas for sugar sweetened beverage was at action/maintenance stage during pretest. (Table 2).

Table 1 Demographic data of fourth-grade subjects, n(%)

	Total Subjects (n=35)	Group A (n=16)	Group B (n=19)
Gender			
Male	11(31.4)	4(25.0)	7(36.8)
Female	24(68.6)	12(75.0)	12(63.2)
Cooking Experience			
No	10(28.6)	3(18.8)	7(36.8)
Yes	25(71.4)	13(81.3)	12(63.2)
Cooking Interest			
No	1(2.9)	0(0.0)	1(5.3)
Yes	34(97.1)	16(47.1)	18(94.7)

Table 2 Subjects' stage of behavioral change during baseline (pretest), n (%)

	Total subjects (n=35)			Group A (n=16)			Group B (n=19)		
	PC	C/P	A/M	PC	C/P	A/M	PC	C/P	A/M
Vegetables	18(51.4)	9(25.7)	8(22.9)	6(37.5)	4(25.0)	6(37.5)	12(63.2)	5(26.3)	2(10.5)
Fruits	19(54.3)	6(17.1)	10(28.6)	6(37.5)	3(18.8)	7(43.8)	13(68.4)	3(15.8)	3(15.8)
Sugar Sweetened Beverages	11(31.4)	9(25.7)	15(42.9)	3(18.8)	5(31.3)	8(50.0)	8(42.1)	4(21.1)	7(36.8)

PC indicates precontemplation or do not plan to change; C/P indicates contemplation/preparation or plan to change; A/M indicates action/maintenance or have changed unhealthy behavior.

Nutrition Knowledge (NK) Subjects in Group A which attend cooking class first increased the median score for nutrition knowledge by 1.5 ($p<.05$) during midterm test whereas subjects in Group B which attend nutrition education class first showed and increased by 3.5 ($p<.05$). (Table 3). Both Group A and Group B showed significantly increase in median score during posttest compared to pretest. It was also noticed that median score for both Group A and B during final test, showed a significantly increase ($p<.05$) compared to pretest. Subjects in Group B who attend nutrition education curriculum first then experiential cooking showed a maintenance in median score at 10 since midterm test, however subjects in Group A who attend experiential cooking curriculum first then nutrition education showed a significant decreased of median score by 1 ($p<.05$) during the final test.

Selecting Attitude (AT) Attitude score for subjects in Group B showed a significantly higher than Group A by 2.5 ($p<.05$) during the final test (Table 3). After the whole program, both Groups showed a decrease in attitude score, but it did not show statistically significant. Median score for selecting attitude of subjects within both groups considered the same during pretest and final test.

Consuming Behavior (BE) The median of Group A subjects' behavior towards FV and SSB doesnot show significantly increase in neither midterm test nor posttest compared to pretest, yet it showed a significantly increased by 4.5 ($p<.05$) during follow-up test. However, median of Group B subjects showed a significant increased by 2 and 2.5 ($p<.05$) during midterm test and posttest respectively, but there was no statistically different between midterm test and posttest.

Table 3 Nutrition knowledge, selecting attitude and consuming behavior score of vegetables, fruits and sugar sweetened beverages during pretest, midterm test, posttest, follow-up test and final test, median (IQR)

	Group A (n=16)[1]					Group B (n=19)[1]				
	W_0 (Pretest) (Before intervention)	W_4 (Midterm Test) (After cooking)	W_8 (Posttest) (Cook-Education)	W_{10} (Follow-up Test (2 weeks after program)	W_{20} (Final Test) (After winter break)	W_0 (Pretest) (Before intervention)	W_4 (Midterm Test) (After Education)	W_8 (Posttest) (Education-Cook)	W_{10} (Follow-up Test (2 weeks after program)	W_{20} (Final Test) (After winter break)
Nutrition Knowledge [2]	6.5 (5-8)[a]	8 (7-10.75)[b]	10.5 (9-11)[bc]	10 (9-11)[c]	9 (8-10)[b]	6.5 (5-8)[a]	10 (8-11.25)[b]	10 (9-12)[b]	10 (9-12.25)[b]	10 (9-11)[b]
Selecting Attitude [2]	52 (49.25-54.75)	50.5 (48-54)	50.5 (46.25-56.5)	52 (43.25-55)	52* (43.25-55)	54.5 (49.5-57.25)	54 (49-56)	53 (46-56.25)	52 (47.75-55.25)	54.5* (49.25-57)
Consuming Behavior [2]	46.5[a] (34.25-54.25)	49[a] (33-54.75)	48 (37.5-56.75)	51[b] (35.25-58.5)	48 (38.5-57.25)	43[a] (33.75-53.5)	45[b] (38.5-58.25)	45.5[b] (41.25-56.25)	47 (40-53)	46.5 (38.75-58.25)

1. Median in the same row with * marks are significantly different ($p<.05$)
2. Median in the same row with different letters are significantly different ($p<.05$)

Score range: Nutrition Knowledge 0-15; Selecting Attitude 12-60; Consuming Behavior 12-60

Stages of Behavior Change Behavior change in this study indicated that subjects consumed two daily servings of vegetables, one daily serving of fruits and less than four times per week of sugar- sweetened beverages. Regress indicates backward movement to an earlier stage, stable indicates no change in stage, while progress indicates forward movement to a later stage. Data in Table 4 represents the percentages of subjects evidencing each pattern in experiential cooking curriculum, nutrition education curriculum and the overall intervention. In terms of vegetables and fruits, percentage of progress was more desirable in cooking curriculum than that of nutrition education curriculum which were 20.0% and 25.71% respectively. However, it shows a contrast in terms of sugar sweetened beverage. Percentage of progress was more favorable in nutrition education curriculum (25.71%). As an overall intervention which including both cooking and nutrition education intervention, all subjects either in Group A or Group B, showed a better outcome which progressed to a later stage by 20.0%, 25.71% and 37.14% for vegetables, fruits and sugar sweetened beverages respectively as shown in Table 4.

Table 4 Patterns of stability and change in stage after cooking curriculum, nutrition education curriculum and overall intervention, n (%)

	Cooking Curriculum (n=35) W_4-W_0(A)+W_8-W_4(B)			Nutrition Education Curriculum (n=35) W_8-W_4(A)+ W_4-W_0(B)			Overall Intervention (n=35) W_8-W_0(A)+W_8-W_0(B)		
	Regress	Stable	Progress	Regress	Stable	Progress	Regress	Stable	Progress
Vegetables	3 (8.57)	25 (71.43)	7 (20.0)	8 (22.86)	24 (68.57)	3 (8.57)	3 (8.57)	25 (71.43)	7 (20.0)
Fruits	1 (2.86)	25 (71.43)	9 (25.71)	4 (11.43)	29 (82.86)	2 (5.71)	1 (2.86)	25 (71.43)	9 (25.71)
Sugar-Sweetened Beverages	6 (17.14)	22 (62.86)	7 (20.0)	4 (11.43)	22 (62.86)	9 (25.71)	5 (14.29)	17 (48.57)	13 (37.14)

W_0, Pretest (before intervention); W_4, Midterm test (after first period/4 weeks of curriculum); W_8, Posttest (after second period/4 weeks of curriculum)

DISCUSSION

This study examined the effectiveness of experiential cooking and nutrition education intervention through NK, AT and BE towards FV and SSB. Besides, readiness of subjects to consume two daily servings of vegetables, one daily serving of fruits and less than four times per week of sugar sweetened beverages were assessed by stages of behavior change (SOC) among Fourth-grade elementary school students. Results showed a significant increase in both subjects' nutrition knowledge after cooking program in Group A and after experiential learning in Group B during the first 4-week curriculum. These results are consistent with a meta-analysis of a wide variety of school-based nutrition education programs found that experiential learning programs such as hands-on cooking or gardening can improve nutrition knowledge and vegetable consumption in elementary school children (22). Even though the knowledge score of both Group A and Group B subjects were not significantly improved after the second 4-week curriculum, but both groups of knowledge score during posttest was significantly higher than during pretest. This may be explained by the retention of knowledge gained during the first 4-weeks when Group B received nutrition education intervention before crossover took place, which in crossover design is referred to as a carryover effect (23). Carry-over effect in this situation explained that the subjects from both groups have obtained nutrition knowledge during the first 4-weeks of curriculum. Then, the knowledge obtained was carried forward to the second 4-weeks of curriculum. In addition, knowledge score was able to retain for two weeks and up to ten weeks in Group A and Group B subjects, respectively.

In terms of selecting attitude towards FV and SSB by subjects, no significant difference in attitude score was noticed by both groups after 8- weeks intervention. This might be due to the duration of both cooking and nutrition education programs that could influence children's food-related preferences (24). Median duration of most nutrition education intervention was found to be 10 sessions with duration of 90 minutes. Due to time constraint consideration, the duration in this study was only held for 4 weeks each with a duration of 30 minutes each week. It was relatively low compared to most studies. Subjects' diet -related behavior reported as significant higher immediately after 4-weeks of curriculum and after the whole intervention which was another 4- weeks after. This phenomenon was noticed in Group B subjects which started with nutrition education curriculum first and then cooking curriculum. Even though there was no instant significant increase in behavior score for Group A subjects, yet a significant higher in behavior score was noticed during follow-up test (2-weeks after the intervention) compared to pretest and midterm test. This might show that whenever there was experiential and contextual knowledge program, a lasting impact on diet-related behavior could be noticed (25).

Behavior change in consuming FV and SSB was assessed further through stages of change. No comparison of food consumption frequency between different stages of change was made. Subjects were compared individually beginning from pretest to posttest to obtain the pattern of stability and change in stage after different curriculum. Despite review demonstrated that stage-tailored intervention is preferable to increase the effectiveness and success of nutrition education intervention, nonetheless, this study reported that the whole program which included both cooking and nutrition education curriculum could facilitate subjects' behavior change to progress to a later stage rather than only one type of curriculum. The results also indicated that different curriculum might be suitable for different food items. For instance, the effect of cooking curriculum was found to be more suitable for vegetables and fruits while nutrition education curriculum which mainly carried out through lecturing, drama in education, and games were more effective for sugar sweetened beverages. Surprisingly, as the result suggested, a combination of both cooking and nutrition education curriculum would eventually provide a better outcome in terms of facilitating subjects to move forward to a later stage which lastly changed their unhealthy behavior especially for sugar sweetened beverages.

CONCLUSION

The findings of this study demonstrate that both the cooking curriculum and the nutrition education curriculum independently contributed to significant improvements in nutrition knowledge and dietary behaviors among elementary school children, particularly in relation to fruit and vegetable (FV) intake and reduced consumption of sugar-sweetened beverages (SSB). Importantly, the most compelling outcome emerged when both curricula were implemented together. The integration of practical cooking skills with structured nutrition education produced a synergistic effect, reinforcing knowledge while simultaneously enabling children to apply it in real-life contexts. This dual approach facilitated progression to more advanced stages of behavioral change, suggesting that children were not only learning but also internalizing and practicing healthier habits.

Such progression is critical, as it reflects the potential for sustained lifestyle modifications rather than

short-term improvements. By combining experiential learning with theoretical understanding, the intervention empowers children to make informed food choices and actively engage in their own health management. This integrated model is particularly effective in the elementary school setting, where early interventions can establish foundational habits that persist into adolescence and adulthood.

In conclusion, the combined cooking and nutrition education curricula provide a comprehensive framework for promoting healthy eating concepts in schools. Beyond improving knowledge and immediate behaviors, this approach supports long-term behavioral change, offering valuable insights for educators, policymakers, and health professionals seeking to design impactful school-based interventions. The evidence underscores the importance of holistic strategies that address both knowledge and practice in fostering lifelong healthy eating behaviors.

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Report

The Impact and Current Situation of Silent Eating during School Lunches in Aomori Prefecture, Japan, during the COVID-19 Pandemic

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ABSTRACT *Background and purpose.* This study examined the current situation of silent eating during school lunchtime in Aomori Prefecture, Japan, within the context of the globally impactful COVID-19 pandemic, focusing on the period after COVID-19-related restrictions were lifted. *Methods.* In August 2024, a self-administered questionnaire survey was distributed to children and class teachers at four public elementary schools. The children's perception of conversation during school lunchtime was assessed through a questionnaire. Class teachers were surveyed about the lunch location, desk arrangement, and silent eating guidance. This study examines the current situation during the normality after the COVID-19 pandemic and compares the findings with previous research. *Result.* Regarding "Conversation during Lunch," 66.7% of respondents reported that they "always" or "sometimes" engage in conversation during lunch. Regarding "Desire to Talk while Eating," 73.3% agreed with the statement. For "Enjoyment of Eating while Talking," 81.7% agreed. These findings show an increase in specific items compared to previous research. *Conclusion.* Revealed that conversation during lunchtime has increased, and the impact of the silent eating policy during the COVID-19 pandemic appears to have diminished significantly. Additionally, the need to enhance 'Social Skills' through food education for children who experienced the pandemic was identified.
Keywords: COVID-19, silent eating, children, school lunch, *shokuiku* (Food and Nutrition Education)

INTRODUCTION

I. Premise

1. School Lunch - The Nutritional and Educational Aspects of Japanese School Lunch -

1) Nutritional Standards in Japanese and Foreign School Lunch

In Japan, school lunch is planned and provided based on the school lunch Implementation Standards (1). This ensures that children receive a balanced intake of necessary nutrients. This framework is designed to ensure that school lunches provide children with the nutrients they need. In contrast, various countries have set nutritional standards and criteria for selecting ingredients in school lunch. The goal is to offer meals that meet certain nutritional requirements (2).

2) Food and Nutrition Education in Japan and Other Countries

In Japan, school lunches are not only about providing food but are also integrated into the educational system. The concept of "*Shokuiku*" (Food and Nutrition Education) has spread, where education is delivered through school lunch. Conversely, in many foreign countries, school lunch programs are more focused on nutritional supplementation. The educational aspect of Food and Nutrition Education is often not as emphasized as it is in Japan. While many foreign countries also promote measures for nutritional balance and health improvement, Food and Nutrition Education activities are not as systematically structured as in Japan (3).

3) Positioning of Food and Nutrition Education in Japanese School Lunch

In Japan, the Basic Law on Food and Nutrition Education was enacted in 2005. It positions Food and Nutrition Education as "fundamental to life and the foundation of intellectual, moral, and physical education." Thus, Japanese school lunch is expected to not only ensure adequate nutrition but also fulfill an educational role, including Food and Nutrition Education (4). While Food and Nutrition Education is conducted as part of the curriculum, mealtime also serves to foster social skills. Children learn to interact by sharing meals together in the lunchroom regardless of grade level. They also develop social skills by sitting face-to-face with classmates in the classroom.

II. Introduction

The impact of the COVID-19 pandemic (hereafter, 'the pandemic') has been reported to include changes in

children's nutrition and dietary habits. During the state of emergency, a decline in the consumption of nutritionally balanced meals was observed, particularly among children from lower-income households (5). It has been revealed that children with delayed wake-up and breakfast times during school closures were found to be at higher risk of developing unhealthy lifestyle habits (6). Furthermore, after schools reopened, intake levels of thiamine, vitamin B6, potassium, fruits, and dairy products increased. Meanwhile, consumption of sugars, sweets, and sugary drinks decreased (7). These findings suggest that the pandemic, especially during the state of emergency and school closures, had a significant impact on children's nutritional status and eating habits.

The pandemic also affected Food and Nutrition Education initiatives. The "*Guidelines for Food and Nutrition Education*," developed to enhance the promotion of Food and Nutrition Education in schools, outline six key perspectives for food-related instruction (8). Among them, the "Social Skills perspective (developing dining manners and interpersonal relationship-building skills through meals)" emphasizes the value of mealtime as an important opportunity for social interaction. It also highlights fostering an attitude of active engagement in communication.

In relation to children, however, the Ministry of Education, Culture, Sports, Science and Technology (MEXT) issued a notification on November 22, 2021, titled "*Hygiene Management Manual for COVID-19 in Schools*" (9). It stipulated that during lunchtime in school cafeterias, students should avoid siting facie-to-face, refrain from speaking loudly, and eat alone. Consequently, the social function of mealtime as an opportunity for communication was significantly disrupted.

The "*Basic Policy for Countermeasures against COVID-19*" issued on November 25, 2022, removed the reference to "silent eating"(10). MEXT then notified local education boards that conversations during school lunchtime could be permitted, provided that appropriate infection control measures in place were implemented (11). Subsequently, on May 11, 2023, COVID-19 was officially reclassified from a “Novel Influenza Infection, etc.” (equivalent to a Category II Infectious Disease) to a Category V Infectious Disease. Following this reclassification, only standard preventive measures—such as handwashing—were required unless an outbreak occurred (12).

Approximately two months after this transition, a study on school lunch revealed that many children perceived eating while conversing to be enjoyable. However, a noticeable reluctance to initiate conversation remained (13).This suggests that the psychological and behavioral effects of prolonged "silent eating" policies may persist even after the formal restrictions have been lifted.

Given these circumstances, it is essential to evaluate the long-term impact of silent eating policies more than one year after COVID-19 was classified as a " Class V Infectious Disease." Such evaluation is expected to contribute to more effective support strategies through food-related instruction provided during school lunch programs.

Based on this premise, the present study examines the current situation in Aomori Prefecture, Japan, a specific region. This description is given in the context of the globally impactful COVID-19 pandemic, focusing on the situation following the end of the normality after the COVID-19 pandemic. Additionally, changes in the situation from the period during the pandemic and normality after are discussed by comparing the current findings with those from previous research (13).

MATERIALS AND METHODS

1. D

1. Data for the study

(b) Compare

			(a) This study			
	Previous research (13)			Intervention study		
Months and Years	Jul. 2022	Jul. 2023	Aug. 2024	Sep. 2024	Oct. 2024	Dec. 2024
Method			Baseline survey	Post survey	One-month post-survey	Three-month post-survey
	Self-administered questionnaire		Self-administered questionnaire			

Target Schools	Six elementary schools in Aomori Prefecture (A, B, C, D, E, F)	Four elementary schools in Aomori Prefecture (A, B, C, D)

Fig 1. Summary Chart of Survey Data for this study

The data used for analysis in this study were cross-sectional observational data obtained from an intervention study ((Figure 1 (a)). The intervention study was conducted at the same elementary school as in previous research (13). The intervention study aimed to examine whether a school broadcasting system could be effectively utilized for nutrition education in elementary schools. It also aimed to explore its impact on students' conversations during lunchtime. A baseline survey, consisting of a self-administered questionnaire, was conducted in August 2024 (baseline survey). Following this, a one-month school broadcasting program was implemented in September 2024. After the program concluded, a post-program survey was distributed, and responses were collected. Subsequent surveys were conducted one month and three months after the completion of the broadcasting program. For the purpose of this study, the observational data obtained from the baseline survey were used as the primary outcome.

2. Characteristics of the Survey Area (14)

Aomori Prefecture is located in the northern part of Japan. It is bordered by the Pacific Ocean to the east and the Sea of Japan to the west. It has a cool climate characterized by short summers and long winters. The population of Aomori is approximately 1.2 million, which is smaller compared to other Japanese prefectures, and over 30% of the population is aged 65 or older, indicating an aging population. Its geographical features include being separated from Hokkaido to the north by the Tsugaru Strait, bordered by the Pacific Ocean to the east, and the Sea of Japan to the west. To the south Aomori shares borders on Akita and Iwate Prefectures. The total area of Aomori is 9,646 km² (the eighth largest in Japan), accounting for 2.6% of the country's total land area. However, the population density ranks 41st nationally, placing it in a region with vast, rich natural environments. The western part of the prefecture, benefiting from favorable temperatures and sunlight, is known for apple production, while the eastern region, taking advantage of its cool summer conditions, is active in vegetable farming and livestock production. As a result, local ingredients are actively used in school lunches, with the proportion of locally sourced products (based on monetary value) at 66.9%, which is higher than in other Japanese prefectures (15).

3. Participants

This study was conducted with the cooperation of six public elementary schools in Aomori Prefecture. Of these, four schools agreed to cooperate with the survey. The participants consisted total number of 150 children from grades 4, 5, and 6 enrolled in these four schools. Consent for participation was obtained from both the children’s parents and the children themselves, resulting in 120 participants (61 boys and 59 girls). The remaining 30 participants either declined to participate or did not return the consent forms. Thus, the final sample size for the study was 120. Additionally, to understand the characteristics of the target schools, a self-administered questionnaire survey was conducted with eight class teachers.

4. Questionnaires

The scales for each question were set respectively based on previous studies (13).

To describe the conversation situation during school lunchtime, the question "Do you talk during school lunchtime?" was asked, with responses ranging from "Always" to "Never" on a four-point scale.

For the other related item, " Do you eat all your school lunch?" responses were collected on a three-point scale, ranging from "Always finish their meal " to " Always leave leftovers."

For the question "Is school lunchtime enjoyable?" responses were collected on a four-point scale, ranging from "Very enjoyable" to "Not enjoyable."

For the questions "Do you want to eat school lunch while talking with others?" "Do you think it is fun to eat school lunch while talking with others?" and "Do you think it is possible for everyone to eat school lunch while talking?" responses were collected on a four-point scale, ranging from "Agree" to "Disagree."

Additionally, class teachers responded to items regarding the lunch location, desk arrangement, and silent eating guidance.

5. Statistical analysis

The analysis was conducted on data from 120 participants who provided informed consent, including both the children and their guardians. Missing data were handled using listwise deletion on a per-item basis. To examine differences between male and female participants for each item, the Mann–Whitney U test was

employed for statistical comparisons. The analysis software used was EZR (16) Version 1.55 (Department of Hematology, Saitama Medical Center, Jichi Medical University), and the significance level was set at and the significance level was p-value < 0.05.

The results obtained in this study were compared with those from previous research (13) conducted by the authors (Figure 1 (b)). The previous studies, which targeted fifth-grade children from six elementary schools (A, B, C, D, E, F) in July 2022 and July 2023, were also conducted in these schools. In the present study, surveys were conducted in August 2024, targeting fourth, fifth, and sixth-grade children from four of the same elementary schools (A, B, C, D), where consent had been obtained.

The term "July 2022" in the text refers to the period during the seventh wave of the pandemic in Japan. During this time, the number of new cases exceeded previous waves (17). This period indicates the peak of the so-called "during the COVID-19 pandemic." "July 2023" refers to the situation approximately two months after the classification of COVID-19 was changed. The classification was changed from "Novel Influenza Infection, etc. (Equivalent to Class II Infectious Diseases)" to "Class V Infectious Disease" (effective from May 8, 2023) (18). This change represents the "after the COVID-19 pandemic" phase of the pandemic. "August 2024" refers to a period more than a year after COVID-19 was categorized as a "Class V Infectious Disease." This period marks the " normality after the COVID-19 pandemic," with no restrictions in place.

6. Ethical considerations

This study was conducted with the review and approval of the Ethics Committee of Aomori University of Health and Welfare (Approval No. 24047). The class teachers verbally explained the purpose, methods, and the voluntary nature of participation in the survey to the children. They ensured that there would be no disadvantage for those who chose not to participate. The parents were provided with written information detailing the research outline and ethical considerations. Written consent was obtained from both the parents and the children before the survey began.

RESULTS

1. Characteristics of the Target Elementary Schools (Table 1)

Table 1. Status of School Lunches at Target Elementary Schools

		Overall (n=8)	
		n	(%)
Lunch Location	Classroom	7	87.5
	Lunchroom	1	12.5
	Other	0	0.0
Desk Arrangement	Facing Each Other (Group Format)	1	12.5
	Same Direction	7	87.5
	Other	0	0.0
Silent Eating Guidance	Yes	4	50.0
	No	4	50.0

Regarding the lunch location, most children ate in their classrooms. Some schools had a designated lunchroom. As for the desk arrangement, most of the classrooms had children seated facing the same direction, but some were arranged in facing each other (group format). Regarding silent eating guidance, responses were evenly split between those who followed this practice and those who did not.

3. Conversation and Eating Behavior during School Lunchtime of children (Table 2)

Table 2. Distribution of Conversation and Eating Behavior during School Lunchtime of children

		Overall		Boys (n=61)		Girls (n=59)		
		n	(%)	n	(%)	n	(%)	*p*-value
Frequency of Food Leftover	Always finish their meal	64	53.3	35	57.4	29	49.2	0.452
	Sometimes leave leftovers	49	40.8	22	36.1	27	45.8	
	Always leave leftovers	7	5.8	4	6.6	3	5.1	
Conversation during Lunch	Always	38	31.7	21	34.4	17	28.8	0.619
	Sometimes	42	35.0	21	34.4	21	35.6	
	Rarely	33	27.5	15	24.6	18	30.5	
	Never	7	5.8	4	6.6	3	5.1	
Enjoyment of Lunchtime	Very enjoyable	50	41.7	26	42.6	24	40.7	0.988
	Moderately enjoyable	60	50.0	29	47.5	31	52.5	
	Not very enjoyable	9	7.5	6	9.8	3	5.1	
	Not enjoyable at all	1	0.8	0	0.0	1	1.7	
Desire to Talk while Eating	Agree	88	73.3	46	75.4	42	71.2	0.654
	Somewhat agree	25	20.8	11	18.0	14	23.7	
	Somewhat disagree	6	5.0	4	6.6	2	3.4	
	Disagree	1	0.8	0	0.0	1	1.7	
Enjoyment of Eating while Talking	Agree	98	81.7	52	85.2	46	78.0	0.291
	Somewhat agree	16	13.3	7	11.5	9	15.3	
	Somewhat disagree	6	5.0	2	3.3	4	6.8	
	Disagree	0	0.0	0	0.0	0	0.0	
Ability to Eat while Talking	Agree	67	55.8	39	63.9	28	47.5	0.106
	Somewhat agree	28	23.3	11	18.0	17	28.8	
	Somewhat disagree	21	17.5	9	14.8	12	20.3	
	Disagree	4	3.3	2	3.3	2	3.4	

Mann-Whitney U test

Each item was compared between genders. The results showed no significant differences between males and females for any item, and thus, subsequent analyses were conducted with both genders combined.

Regarding the " Conversation during Lunch," 66.7% of respondents answered that they "always" and "sometimes" engage in conversation. Regarding " Desire to Talk while Eating," 73.3% answered "Agree." When asked, " Enjoyment of Eating while Talking " 81.7% answered "Agree."

DISCUSSION

1. Changes across Three Time Points (during the COVID-19 Pandemic, after the COVID-19 Pandemic, and Normality after the COVID-19 pandemic)

Notifications Related to Infection Control		Months and Years	Conversation during Lunch (%)	Target Schools	Target Children's (Grade)	Social Situation at the Time of the Survey	The COVID-19 Pandemic
Response to Eating Alone	→	Nov. 2021					Pandemic
		Jul. 2022	21.5	Six elementary schools in Aomori Prefecture (A, B, C, D, E, F)	5th *Recalling 4th	The period during the 7th wave, when the number of new positive cases surpassed previous waves in Japan.	
Removal of Silent Eating Guidelines	→	Nov. 2022					
Routine Infection Control Measures	→	May 2023					
		Jul. 2023	38.3	Six elementary schools in Aomori Prefecture (A, B, C, D, E, F)	5th	The period approximately two months after the COVID-19 was reclassified as a "Class V Infectious Disease" in Japan.	Post-pandemic
		Aug. 2024	66.7	Four elementary schools in Aomori Prefecture (A, B, C, D)	4, 5, 6th	The period more than one year after the COVID-19 was reclassified as a "Class V Infectious Disease" in Japan.	Normality

"Conversation during Lunch (%)", answered "always" and "sometimes."

Fig 2. Comparison of Conversation during School Lunchtime in Previous Studies and the Present Study

After the COVID-19 pandemic, a notable increase was observed in conversation during school lunchtime (Figure 2) among the children. More children who found eating while talking enjoyable were also observed. Additionally, there was an increase in the number of children who wished to talk while eating. These changes indicate a gradual return to pre-pandemic norms, when silent eating was not enforced.

Compared with previous research (13), during and after the pandemic, children ate in their classrooms facing the same direction. Furthermore, during the pandemic, silent eating was enforced in all cases. In

comparison with previous research (13), the results for "Conversation during Lunch" at three different time points were as follows. During the pandemic, 21.5% answered "always" and "sometimes." After the pandemic, 38.3% responded this way. During the normality after the COVID-19 pandemic, the rate rose to 66.7%.

Regarding " Desire to Talk while Eating," 57.0% responded "Agree" during the pandemic, 60.7% after the pandemic, and 73.3% during the normality after pandemic phase. For " Enjoyment of Eating while Talking ", 75.5% responded "Agree" during the pandemic, 77.6% after the pandemic, and 81.7% during the normality after pandemic phase.

Previous research (13) indicated that although children desired to talk while eating after the pandemic, few initiated conversations themselves. The effects of silent eating due to the COVID-19 pandemic still persisted. However, in the normality after the COVID-19 pandemic, approximately 70% of children wanted to talk while eating, and around 70% engaged in conversation during school lunchtime. This suggests that the gap between the desire to talk while eating and the ability to do so has narrowed. In other words, the effects of silent eating during the COVID-19 pandemic seem to have diminished in the normality after the COVID-19 pandemic. In elementary schools, the practice of silent eating has been gradually discontinued, and school lunchtime settings such as lunchrooms and Facing Each Other (Group Format) have become more common. On the other hand, children who experienced the pandemic may lack certain "Social Skills," including developing dining manners and interpersonal relationship-building skills through meals. These challenges, highlighted from a food and nutrition education perspective, may stem from the prolonged period of restricted interaction. This suggests a need for further development of the understanding that mealtime is an important opportunity for communication and fostering positive attitudes that encourage active communication during meals.

This report includes inferences based on the temporal context of the pandemic. It provides new insights into the post-pandemic period, particularly following the lifting of restrictions on conversation during school lunches. There are few reports that focus on the situation after such restrictions were lifted, making this finding a novel contribution.

There are three primary limitations in this study. First, since the research was conducted in a limited region of Aomori Prefecture, caution is needed when generalizing the results. To generalize the findings, it will be necessary to investigate whether similar trends are observed in other regions. Second, it is possible that Aomori Prefecture, with its relatively low population density, experienced a lesser degree of the COVID-19 impact compared to other regions of Japan. Third, the study did not compare exactly the same group of children. Although the research was conducted in the same schools as the previous study, two schools were excluded. The children sample included a broader range (grades 4, 5, and 6).

While the groups were not identical, capturing changes across three time points in response to silent eating provides significant insights. Despite these limitations, this study clarifies the current situation, where the impact of silent eating imposed during the COVID-19 pandemic has substantially diminished. It also contributes to the generation of new hypotheses for future strategies to support children. This applies to the normality after the COVID-19 pandemic, after the removal of restrictions.

In conclusion, it has been revealed that conversation during school lunchtime has increased. The impact of the silent eating policy enforced during the COVID-19 pandemic appears to have diminished significantly. On the other hand, the findings suggest the continued need for further enhancement of "Social Skills" from the perspective of food education for children who experienced the pandemic. This study has provided valuable implication to develop the post-pandemic strategies for supporting school lunch programs.

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