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Original Article

The Development of Wax Cubes for Chewing Ability Evaluation : A Preliminary Study and its Clinical Application

Kittima Prapatrungsri
Dental Department
Banphaco Hospital
Faculty of Dentistry, Chulalongkorn University
Amorn Petsom
Assistant Professor
Department of Chemistry
Faculty of Science, Chulalongkorn University
Orapin Kaewplung
Associated Professor
Department of Prosthodontic
Faculty of Dentistry, Chulalongkorn University

Correspondence to:
Dr. Orapin Kaewplung
Department of Prosthodontic
Faculty of Dentistry Chulalongkorn University
Henri-Dunant Rd., Patumwan Bangkok 10330
Tel: 02-2188532-3
Fax: 02-2188534
E-mail: a_ae_aea@hotmail.com,
orapin.ka@chula.ac.th

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Abstract

The purpose of this study was to develop wax cubes having the desired physical properties, for evaluating the chewing ability. The manufacturing and evaluating process can be done in a short time. The developed wax cubes can be used to evaluate the chewing ability among various subject age groups. Four types of wax were blended until they achieved the desired physical properties to create 10 mm. x 10 mm. x 10 mm. wax cubes. Forty-five subjects (21 males and 24 females) with normal occlusion were classified into 3 groups (Group 1: 20-29 years old, Group 2: 30-39 years old, and Group 3: 40-49 years old). The wax cubes were disinfected and stored in an incubator (37°C) for 24 hours then soaked in a water bath (37°C) for 10 minutes prior to testing. Each subject was assigned to chew four pieces of the wax cubes, one by one for ten chewing strokes each using habitual chewing patterns. The chewed wax was captured and analyzed by the Image J program (NIH) which calculated the percent of well mixed color areas. The results showed the average percentage of chewing ability based on well mixed areas in Group 1, 2 and 3 were 25.3±4.4, 23.6±5.7, and 16.5±5.5 (mean±s.d.), respectively. One-way ANOVA showed the differences between Group 1 and 3, Group 2 and 3 were statistical significance ($p < .05$). However, no statistically significance ($p > .05$) was obtained between gender in each group when analyzed by Independent t-test. In conclusion, the wax cubes developed in this study using an uncomplicated manufacturing and evaluating process can be used to identify chewing ability among different groups. Also, this study indicates chewing ability and age of subjects are negatively correlated.

Key words: chewing ability; color histogram; wax cubes

Introduction

The objectives of dental treatment are to restore natural teeth and the masticatory system to maintain oral function as well as to preserve facial appearance.[1] The effectiveness of dental treatment can be evaluated by various methods, for instance, patient satisfaction and masticatory function; speech, chewing and deglutination. Previous studies showed that loss of chewing ability adversely affected people's quality of life.[2-7] Therefore, the evaluation of chewing ability has become one of the most important

factors for the dental professional to assess the success of a dental treatment.[8]

There are two methods to evaluate the chewing ability. The first method is the subjective evaluation, which is an individual assessment by questionnaire or personal interview. This method has some limitations such as the results that are based on perceptions of the subjects and the lack of exact criteria. Also, the data could not be compared among the subjects.[9-13] Therefore, the objective evaluation, which is a quantitative method, has become more popular,[14,15] with various techniques developed for this method. Initially, the comminuting test was used. This method used natural or synthetic test food to evaluate chewing ability by detecting the amount of particles which could pass through sieves, the so called "Sieve method".[8,16-23] In addition, this method involves many steps from drying, screening, weighing, and analyzing processes. Therefore, it was a complicated and time-consuming method. However, new test foods and evaluation methods have been developed by various researchers, i.e. sugar leakage detection from chewing gum,[24] detection of the change in color-developing chewing gum by either CIE L*a*b color system[25,26] or a software such as Adobe Photoshop Element®.[27] In 2003, Sato and Fueki[28-30] developed the new paraffin wax cubes and used Digital Image Analyzer to evaluate masticatory function by analyzing the picture of chewed test food.

Such methods are not practical in Thailand due to cost of test food and the inability to perform this complicated analytical process domestically. The aim of this study was to develop the wax cubes and compare the physical properties with the gold standard wax, for evaluating the chewing ability, with a manufacturing and evaluating process which can be done in a short time periods. Finally, the developed wax cubes were used to evaluate the chewing ability in various age groups (among subgroup of Thai population).

Materials & Methods

Development of the wax cubes

Four types of wax; Bee wax, Carnauba wax, Paraffin wax and Microcrystalline wax, were blended with different ratios (by weight) until the desired physical properties were achieved: stable solid phase at room temperature (25°C), sufficient softness for chewing at body temperature (37°C), non adherent to natural or artificial teeth, non-toxic, able to be disinfected without affecting other properties, tasteless, and odorless. The blended waxes were melted and divided into two portions; the first portion was mixed with an oil-based red dye (Apocarotenal 20%S, DSM Nutritional Products Ltd., Switzerland) and the second portion remained a white color. The wax bars were made by pouring the molten wax into silicone molds. (Fig. 1a, b, and c) Four pieces of the wax bars were placed alternating in red and white colors to create a 10 mm. x 10 mm. x 50 mm. wax bar (Fig. 1d), then the wax bar was cut to make a 10 mm. x 10 mm. x 5 mm. wax pattern. Next, two wax patterns were combined together after rotating 90 degrees, in order to make an alternating color scheme. From this step, the 10 mm. x 10 mm. x 10 mm. wax cubes were obtained (Fig. 2). The wax cubes were disinfected by exposure to concentrated 350 mg/l ethylene

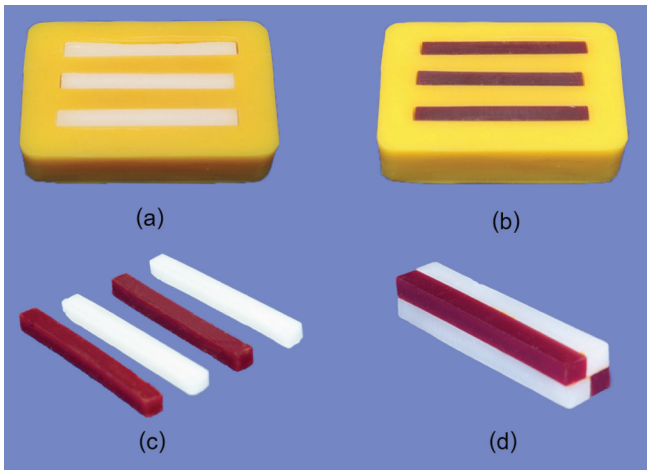

Fig. 1 *The blended wax*
(a) White bar's mold *(b) Red bar's mold*
(c) The wax bars *(d) The 10 mm. x 50 mm. wax bar*

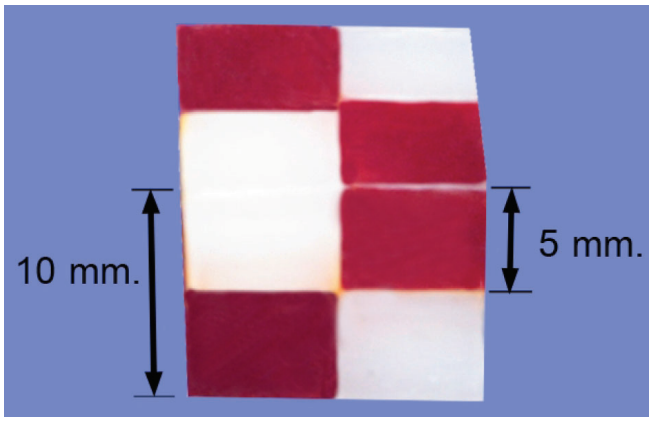

Fig. 2 *The 10 mm. x 10 mm. x 10 mm. wax cube*

oxide gas at 37°C for 5 hours (Steri-Vac4XL, 3M, United States), then the wax cubes were left for 24 hours until the ethylene oxide had vaporized. They were stored at 5°C until used.

Subjects

The study population consisted of 20-49 years old Thais selected to participate based on the following criteria: (1) the subjects had at least 1 premolar and 1 molar per quadrant (occluding pairs were counted as 2 occlusal units); (2) the subjects had no history of neuromuscular disorder, mental disorder, Hepatitis B, Herpes simplex, HIV, and Syphilis. The Ethics Committee of Chulalongkorn University approved the protocol of this study on March 11, 2009 and informed consent for participation in the study was obtained from all subjects. Forty-five subjects (21 males and 24 females) with normal occlusion were classified into 3 groups according to age, 15 persons per each group (7 Male and 8 Female); Group 1: 20-29 years old, Group 2: 30-39 years old, Group 3: 40-49 years old.

Chewing ability test

The wax cubes were brought from the refrigerator and stored in an incubator (Contherm160M, Contherm Scientific Ltd., New Zealand) at 37°C for 24 hours and soaked in a water bath (Isotemp202, Fisher Scientific Co., Ltd, Japan) at 37°C for a further 10 minutes prior to the test. Each subject was requested to sit in an upright position on the dental unit. Lubricant was applied on the occlusal surfaces of the upper and lower posterior teeth. Each subject was told to chew one piece of wax cube on the right side for ten chewing stokes at their habitual position, and duplicate the process again with another wax cube. After finished chewing on the right side, then the subject was told to repeat all of the above chewing process on the left side. Therefore each subject had four pieces of the chewed wax in total. The chewed wax was taken out of the oral cavity, run under tap water for 20 seconds, and soaked in a 70 percent concentration of ethyl-alcohol for 5 minutes before storing in a labeled plastic box.

Chewing ability analysis

Images of the upper and lower sides of the chewed wax were captured by the following devices: (1) a digital camera (Canon EOS 450D) with a macro lens (Canon macro 100 mm.) and a photo stand kit (Copy stand CS920 and Copy light CL-150); (2) 2 light bulbs (Philips® Cool Daylight 125 Watts, Color temperature 6,500 K); (3) a lux meter (DigiconLX-70, Protonics Inter-trade Co, Ltd., Thailand). Every picture was taken under the same conditions: (1) the distance between the chewed wax and the macro lens was 30 cm; (2) the camera was set up as follows; effective pixels = 10 millions, shooting mode as manual, shutter speed = 1/200 sec, aperture value = F8.0, ISO sensitivity = 400 and the picture style was standard. All of the captured images were transferred and analyzed by Image J software (Version 1.42Q, NIH, USA).

The standard color value**,** representing well mixed red and white color wax, was obtained by mixing an equal amount (by weight) of red wax and white wax until a uniform color (orange) of the mixture was achieved (Fig. 3a). Then an image of the mixture was captured and analyzed by the Image J program to define standard color value. The Image J program was used to define color into specific color values; ranging between 0 (white color) to 255 (black color). The program also automatically calculated number of color values, as well as number of pixels, within the defined area. After the analyzing process, Image J program showed the standard color value was in the range of 20-40 (Fig. 3b).

The chewing ability evaluation was done as follows: (1) the pictures of the chewed waxes were analyzed using the measure function of the Image J program to find the total number of pixels on the picture; (2) the picture of chewed waxes were

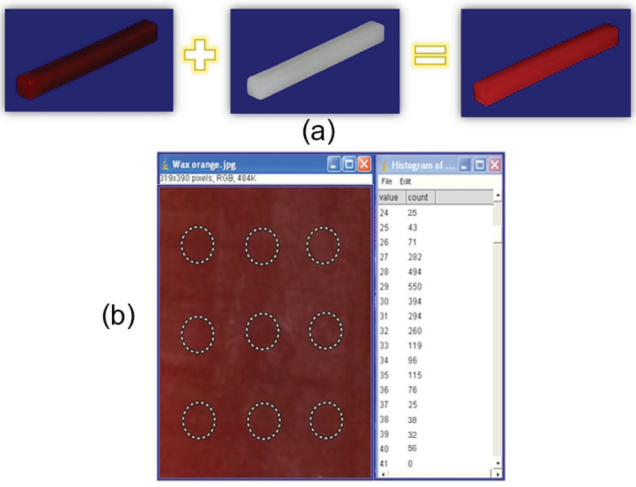

Fig. 3 *Method used to determine the standard color values*
(a) Standard color wax conduction
(b) Standard color values analyzing by Image J program

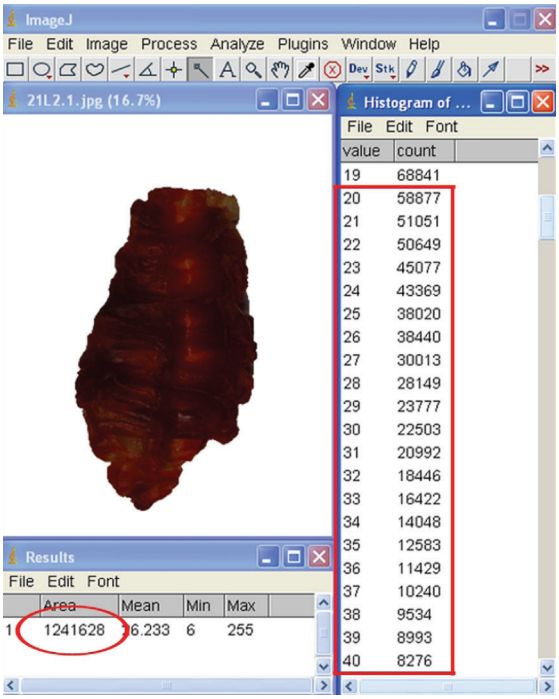

Fig. 4 *Chewing ability evaluation by the Image J program. The red circle represents total area of chewed wax. The red square represents the area having standard color values range between 20-40*

analyzed again using the color histogram function of the Image J program to define the number of pixels which resulted in the standard color values range of between 20-40 (Fig. 4); (3) the percentage chewing ability based on the chewed wax was computed by the following formula: Total number of pixels of standard color value x 100 / Total number of pixels of the chewed wax.

Each subject generated eight surfaces (from four wax cubes) of the chewed wax (Fig. 5), therefore, the average value was calculated in order to determine the average "percentage chewing ability" of each subject.

One-way analysis of variance (ANOVA) and Bonferroni multiple comparison tests were used to compare the results of the three groups ($p < .05$). Independent sample t-test was used to compare the differences among genders in the same age group ($p < .05$).

Results

The developed wax cubes were used to evaluate the chewing ability in three different age groups by using Image J program to analyze the color of the chewed wax.

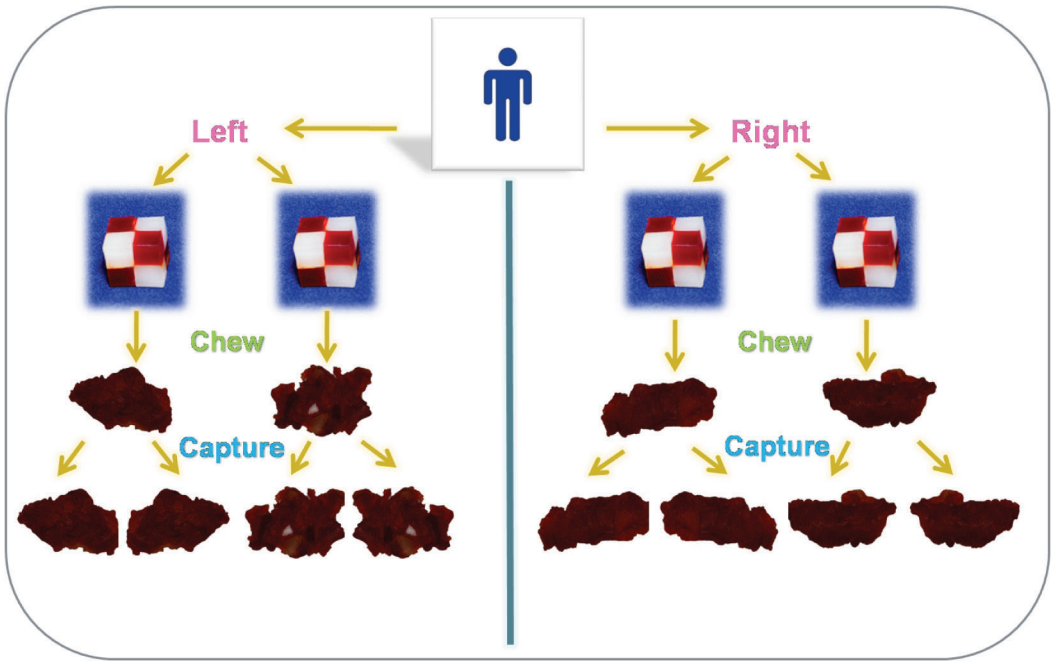

Fig. 5 *The total number of the chewed wax per subject*

The average percentages chewing ability (mean±SD) among the three groups differed as follows: Group 1: 20-29 years old is 25.3±4.4, Group 2: 30-39 years old is 23.6±5.7, Group 3: 40-49 years old is 16.5±5.5. Statistical analyses showed that the data was in a normal distribution with homogeneity variances. One way analysis of variance (ANOVA) revealed a significant difference ($p < .05$) between Group 1 and 3, Group 2 and 3, but had no significant difference between Group 1 and 2 (Table 1).

In the same age group, the average percentage chewing ability between genders was different. Females tend to have a higher average percentage of chewing ability compared with males in all age groups. However, there was no significant difference between gender ($p < .05$).

Table 1 Average percentage chewing

Group	Percentage chewing ability (mean±SD)	
1 (aged 20-29)		25.3±4.4
2 (aged 30-39)	a	23.6±5.7
3 (aged 40-49)		16.5±5.5

"a" denotes statistical difference at alpha = .05

Discussion

The wax cubes developed for this study have physical properties comparable to previous test materials (Table 2). The components of the wax cubes are inexpensive and available in Thailand. The wax cube also has an uncomplicated design, which is easy to manufacture. The physical properties, such as size, shape and texture are easy to standardize. In addition, it is insoluble in water, has no taste as well as no smell, does not stick to natural or artificial teeth, and is soft enough to chew. Chewing the wax can simulate the natural act of chewing real food. The wax cubes are non-toxic because they are produced from edible materials and can be disinfected without any affect on their physical properties. Moreover, the advantage of the symmetric design of the wax cubes allows the subject to chew in any direction with no difference.[28] No subject refused to chew these wax cubes or complained about any discomfort during the test.

Table 2 The comparison of physical properties of Gold standard wax and developed wax

Phsical properties	Gold standard wax	Developed wax
Melting range (°C)	45.0-58.4	41.0-57.4
Phase (at 25°C)	solid	solid
Water solubility	insoluble	insoluble
Water absorption	No	No
Disinfection	-	ethylene oxide gas
Color	2 difference colors	2 difference colors
Material safety	-	approved by the Ethic Committee of Chulalongkorn University

The Image J program was used in this study to evaluate the chewing ability of the subjects. This program is able to define the color value and to calculate the number of pixels in the defined border in an image. Each color has its specific color value that is between 0 and 255 depending on the shade, saturation and brightness of the color. In addition, this program can be downloaded without cost from the Image J program's website (http://rsbweb.nih.gov/ij/) with the permission of the website. Because of its uncomplicated method, the program performs its analysis in just a few minutes and can be used whenever research needs to be conducted.

This study chose the mixing ability test to evaluate chewing ability instead of the comminuting test as it is a better method to distinguish the chewing ability of the subjects. Moreover, this method is simple and requires less time in analysis.[31] Therefore, the chewing ability determined in this study reflects the ability to blend white and red color wax cubes together while chewing in controlled chewing strokes which results from the harmonization of the function between the muscles of mastication, teeth, and periodontium. This study used the color value of well mixed wax, so called "standard color value", to identify the best chewing ability. The chewed wax having more standard color value will represent better chewing ability of those subjects. In this study, the chewing ability of each subject was calculated in an average percentage to compare result among the subjects.

The results of this study showed that the average percentage chewing ability in three age groups were different. It decreased as the age of subject increased; which is consistent with the results of the previous studies.[2,32-34] Moreover, it was found that females tend to have better average percentage chewing ability than males; however, this may be the consequence of the female character which is likely to be that they are able to concentrate more on the assigned task.

This method has many advantages over the sieve method (conventional method) due to its simplicity. The entire processes can be done with a limited budget and time period. As a result, the developed method used here can be utilized clinically to evaluate chewing ability after dental treatment. Comparison of average percentage chewing ability using the gold standard wax cube and the developed wax cubes, as test materials, in same subject group shall be conducted a subsequent study.

Conclusions

The wax cubes used were developed using an uncomplicated manufacturing process and can be used to identify and easily evaluate the chewing ability among selected population subgroups. Also, this study indicates, chewing ability and age of the subjects are inversely correlated, i.e., older subjected were more likely to have a lower percentage of chewing ability.

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บทวิทยาการ

การพัฒนาชิ้นขี้ผึ้งเพื่อประเมินความสามารถในการบดเคี้ยว: การศึกษาเบื้องต้นและการประยุกต์ทางคลินิก

กิตติมา ประภัสร์รังษี
ทันตแพทย์ แผนกทันตกรรม
โรงพยาบาลบ้านแพ้ว (องค์การมหาชน)
คณะทันตแพทยศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย
อมร เพชรสม
รองศาสตราจารย์ ภาควิชาเคมี
คณะวิทยาศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย
อรพินท์ แก้วปลั่ง
ผู้ช่วยศาสตราจารย์ ภาควิชาทันตกรรมประดิษฐ์
คณะทันตแพทยศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย

ติดต่อเกี่ยวกับบทความ:
ทันตแพทย์หญิงอรพินท์ แก้วปลั่ง
ภาควิชาทันตกรรมประดิษฐ์
คณะทันตแพทยศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย
ถนนอังรีดูนังต์ ปทุมวัน กรุงเทพฯ 10330
โทรศัพท์: 02-2188532-3
โทรสาร: 02-2188534
อีเมล: a_ae_aea@hotmail.com, orapin.ka@chula.ac.th

แหล่งเงินทุน: โครงการศูนย์ความเป็นเลิศทางการฟื้นฟูบูรณะช่องปากและใบหน้า

บทคัดย่อ

การศึกษานี้มีวัตถุประสงค์เพื่อพัฒนาชิ้นขี้ผึ้งเพื่อใช้ในการประเมินความสามารถในการบดเคี้ยวให้มีสมบัติทางกายภาพตามที่กำหนด โดยกระบวนการผลิตและการแปลผลสามารถทำได้เองในระยะเวลาสั้น จากนั้นใช้ชิ้นขี้ผึ้งที่พัฒนาขึ้นมาประเมินความสามารถในการบดเคี้ยวในผู้เข้าร่วมวิจัยต่างกลุ่มอายุ ดำเนินการโดยนำขี้ผึ้ง 4 ชนิด มาผสมกันจนได้ขี้ผึ้งที่มีสมบัติทางกายภาพตามต้องการ เพื่อทำชิ้นขี้ผึ้งขนาด 10 มม. X 10 มม. X 10 มม. แบ่งผู้เข้าร่วมวิจัยที่มีการสบฟันปกติ จำนวน 45 คน (ชาย 21 คน หญิง 24 คน) ออกเป็น 3 กลุ่ม (กลุ่มที่ 1 อายุ 20-29 ปี กลุ่มที่ 2 อายุ 30-39 ปี กลุ่มที่ 3 อายุ 40-49 ปี) นำชิ้นขี้ผึ้งไปทำให้ปราศจากเชื้อ เก็บในตู้ควบคุมอุณหภูมิ (37 องศาเซลเซียส) 24 ชม. จากนั้นนำมาแช่ในเครื่องควบคุมอุณหภูมิน้ำ 10 นาที ก่อนเริ่มเคี้ยว ให้ผู้เข้าร่วมวิจัยเคี้ยวชิ้นขี้ผึ้งคนละ 4 ก้อน ครั้งละก้อน ก้อนละ 10 ครั้ง ในตำแหน่งที่ถนัด นำชิ้นขี้ผึ้งที่ผ่านการเคี้ยวแล้วไปถ่ายภาพและประเมินความสามารถในการบดเคี้ยวด้วยโปรแกรมอิมเมจ เจ โดยคำนวณร้อยละของสีที่ผสมกันได้ดี ผลการศึกษาแสดงให้เห็นว่าค่าเฉลี่ยร้อยละ (ค่าเฉลี่ย±ค่าส่วนเบี่ยงเบนมาตรฐาน) ของความสามารถในการบดเคี้ยวใน กลุ่มที่ 1 2 และ 3 เท่ากับ 25.3±4.4, 23.6±5.7 และ 16.5±5.5 ตามลำดับ จากสถิติการวิเคราะห์ความแปรปรวนแบบทางเดียว พบว่ามีความแตกต่างกันอย่างมีนัยสำคัญทางสถิติ ($p < .05$) ระหว่างกลุ่มที่ 1 กับ กลุ่มที่ 3 และกลุ่มที่ 2 กับกลุ่มที่ 3 แต่ไม่พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ ($p > .05$) ระหว่างเพศในแต่ละกลุ่มอายุ เมื่อวิเคราะห์โดยสถิติการทดสอบที โดยสรุป ชิ้นขี้ผึ้งที่พัฒนาขึ้นสามารถแยกความแตกต่างของความสามารถในการบดเคี้ยวระหว่างกลุ่มตัวอย่างทั้ง 3 กลุ่มได้โดยใช้กระบวนการผลิตและการแปลผลที่ไม่ซับซ้อน และพบว่าความสามารถในการบดเคี้ยวมีแนวโน้มลดลงเมื่อกลุ่มตัวอย่างอายุเพิ่มขึ้น