Acta Odontologica Scandinavica, 2009; 67: 113–121

informa
healthcare

ORIGINAL ARTICLE

Analysis of masseter muscle oxygenation and mandibular movement during experimental gum chewing with different hardness

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Abstract

Objective. The purpose of this study was to analyze masseter muscle oxygenation changes and mandibular movements in the experimental chewing of gums with different hardness. ***Material and Methods.*** Subjects for this experiment comprised 23 male volunteers with normal occlusion. Mean age (SD) was 28.3 (2.2) years. Three kinds of gum with mean fracture stresses of 3.52×10^4 N/m^2 (Gum 1), 5.35×10^4 N/m^2 (Gum 2), and 14.0×10^4 N/m^2 (Gum 3) were used. The subjects were instructed to chew gum for 80 s (100 strokes) on the voluntary chewing side at a pace of 1.25 strokes/s. Oxygen saturation in the masseter muscle and mandibular movement during gum chewing were recorded simultaneously using near-infrared spectroscopy tissue oximetry and mandibular kinesiography. ***Results.*** For Gum 1, no subjects showed any significant changes in oxygen saturation during gum chewing. For Gum 2, 10 subjects showed no significant changes, whereas the other 13 showed significant decreases in oxygen saturation. For Gum 3, significant decreases were seen in all subjects. Chewing motions were larger and velocity was higher in gum chewing with decreases in masseter muscle oxygen saturation compared to chewing showing no significant changes. ***Conclusions.*** The results suggest that the harder texture of gum enlarges chewing motion and increases chewing velocity, with an increase in the contribution of anaerobic metabolism to energy yield in masseter muscle. Differences in the responses to gum hardness may indicate individual differences in muscle fatigue tendencies when chewing harder foods.

Key Words: *Gum chewing, masseter muscle oxygen saturation, near-infrared spectroscopy (NIRS), mandibular movements*

Introduction

The relationships between food character, particularly in terms of hardness and chewing movements, have been investigated in many studies. The influences of harder food have been reported as: enlargement of mandibular-opening movements and lateral excursions [1–5], increased velocity of chewing motion [3,6,7], and increased electromyographic activity of the masseter muscle [2,4,8,9].

When relatively hard food is encountered in daily life, feelings of tiredness around the mandibular and difficulty in continuing chewing may arise. Intramuscular blood flow and oxygen saturation are related to fatigue of working muscles [10–12]. However, little is known about blood flow and oxygen saturation changes in masticatory muscles during chewing of food with different hardness.

Recently, near-infrared spectroscopy (NIRS) has been applied to monitor blood flow changes and oxygenation changes in working skeletal muscles [13–22]. Oxygenated and deoxygenated hemoglobin show different light wavelength absorbance, but both absorb light equally at 805 nm. These optical properties of hemoglobin enable continuous and non-invasive monitoring of blood flow and oxygen saturation in muscle [23]. As oxygen supply for muscle is related to blood flow, and changes in oxygen saturation levels are related to metabolism of energy supply for muscle during exercise, the balance between oxygen supply and oxygen requirement and tendencies toward muscle fatigue can be evaluated from NIRS recordings.

As an application of NIRS to the field of dentistry, Delchanho et al. [24] monitored blood flow and oxygen saturation in painful and non-painful human

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(Received 05 September 2008; accepted 30 December 2008)

ISSN 0001-6357 print/ISSN 1502-3850 online
DOI: 10.1080/00016350802715806

masseter muscles and showed that patients with chronic muscle pain display slower intramuscular reperfusion after sustained isometric contraction. Sugisaki et al. [25] reported sex differences in the hemoglobin oxygenation state of resting healthy masseter muscle. Our previous study [26] compared masseter muscle oxygen saturation levels during clenching between normal occlusion and open-bite subjects, and reported that open bite subjects may have problems with muscle fatigue tendency. Besides these, several studies [26–32] have measured blood flow and oxygen saturation in masseter muscle. NIRS could therefore be expected to show differences in blood flow along with oxygen saturation changes in masseter muscle while chewing bolus of varying hardness, helping us better understand the changes in muscle energy metabolism in relation to food hardness.

The purpose of this study was to analyze masseter muscle oxygenation changes and mandibular movements in the experimental chewing of gum with different hardness, and to discuss muscle fatigue induced by chewing harder foods.

Material and methods

Subjects

Subjects for this experiment comprised 23 male volunteers working at Fukuoka Dental College Medical and Dental Hospital. Mean (SD) age was 28.3 (2.2) years (range 25.2–34.6 years). All showed a Class I molar relationship and normal incisor relationships, and none had any symptoms of temporomandibular joint dysfunction or history of chronic muscle pain in the head or neck regions. The experimental protocols were approved by the ethics committee of Fukuoka Dental College, and informed consent was obtained from all subjects.

Experimental condition of gum chewing

Three conditions in gum hardness were set using ordinary commercially available chewing gum (Xylitol; Lotte, Tokyo, Japan), a commercially available harder gum for chewing training (Kamuzougum; Matsuya Jimokuji Center, Aichi, Japan), and a mix of 75% ordinary and 25% harder gum. The experimental conditions of gum hardness were determined as described later. As a physical property of these chewing gums, fracture stress after 3-min ordinary chewing was measured using a creep meter (Model RE2-3305; Yamaden, Tokyo, Japan) in 10 samples of each gum. Mean fracture stress was 3.52×10^4 N/m^2 for the commercial ordinary gum (Gum 1), 5.35×10^4 N/m^2 for the mixture of the two commercial gums (Gum 2), and 14.0×10^4 N/m^2 for the chewing training gum (Gum 3). The weight of each gum was also measured. Mean weights were 1.61 g for Gum 1, 1.64 g for Gum 2, and 1.62 g for Gum 3. One-way analysis of variance with Scheffe's multiple comparison post hoc test indicated significant differences in fracture stress, but no differences in weight among the three kinds of gum.

Measurement of masseter muscle oxygen saturation and mandibular movement

Oxygen saturation in the masseter muscle and mandibular movement during gum chewing were recorded simultaneously (Figure 1). Prior to measurement, the voluntary chewing side was decided by having the subject chew Gum 1 freely for 3 min and asking the subject which side was preferred. Gum 2 and Gum 3 were also freely chewed for 3 min, and the 3 kinds of gum were kept in a small laboratory dish. Muscle oxygen saturation was measured continuously using a 2-wavelength (750.0 nm, 830.0 nm) NIRS Tissue Oximeter (PSA-500; Biomedical Science, Kanazawa, Japan). The NIRS probe was placed in the middle of the masseter muscle antero-posteriorly on the voluntary chewing side, parallel to the main direction of the muscle fibers, as far as possible in a standardized position by palpation of the muscle. The probe was attached with medical double-sided tape on the masseter muscle so that the light emission and detection parts of the probe were positioned across the line connecting the tragus of the ear and the angle of the mouth. Measurement depth was 20 mm from the skin surface, covering both superficial and deep portions. Mandibular movement was measured using a mandibular kinesiograph (K6-I Diagnostic System EX; Myotronics-Noromed, Takwila, Wa.,

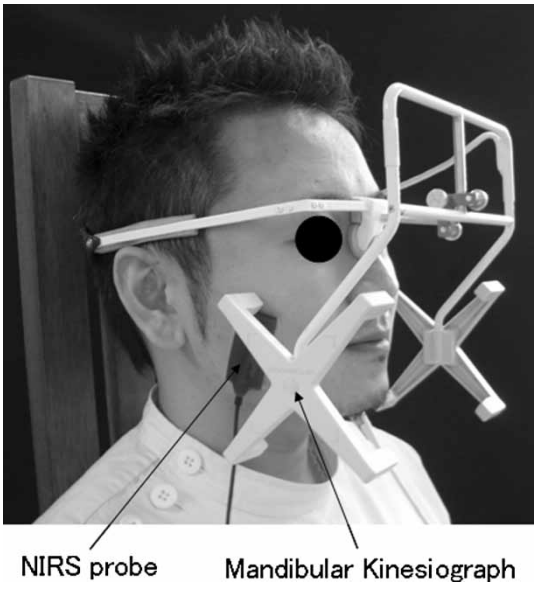

Figure 1. Measurement of masseter muscle oxygen saturation and mandibular movement. The near infrared spectroscopy (NIRS) probe and kinesiograph are mounted on the subject.

USA). Prior to the experiment, absence of perturbations of NIRS and kinesiographic data due to simultaneous recordings had been verified. Each subject was placed in a dental chair so that the Frankfort horizontal plane became parallel to the floor. Frankfort horizontal plane was identified as passing through the lowest point in the floor of the left orbit and the highest points on the margins of the external auditory meati. Subjects were instructed to chew gum for 80 s (100 strokes) on the voluntary chewing side at a physiological pace of 1.25 strokes/s [33] following a metronome. Hemoglobin concentration and oxygen saturation levels were recorded at a sampling frequency of 1 Hz, from 90 s before the start of chewing until 3 min after the end of chewing. The traces of movements of mandibular incisor points were recorded three-dimensionally during gum chewing. The experiment was performed in order from Gum 1 to Gum 3, with a resting interval between each trial of >30 min.

Analysis of oxygen saturation in masseter muscle

To determine significant changes in oxygen saturation for each subject in the three gum chewing conditions, oxygen saturation levels in the 10 s before the start of chewing and in the 10 s before finishing chewing were compared using the Mann-Whitney U-test for non-normally distributed data. Values of $p<0.05$ were regarded as statistically significant.

Analysis of mandibular movements

From recordings of traces of mandibular movements during chewing, the following parameters were measured in each stroke (Figure 2). Traces were zeroed at the origin when the cusps of the upper and lower teeth were in full interdigitation (CO position).

1. Maximum vertical opening from CO
2. Maximum antero-posterior movement from CO
3. Maximum lateral deviation
4. Maximum opening velocity
5. Maximum closing velocity

The mean value at an interval of 30 strokes was calculated for each of the 5 parameters. The interval was set in each subject as described below. To determine significant differences between the three conditions of gum chewing, data were subjected to one-way analysis of variance with Scheffe's multiple comparison post hoc test. Values of $p<0.05$ were regarded as statistically significant.

Results

Changes in masseter muscle oxygen saturation levels during gum chewing

Figure 3 shows typical results of muscle oxygenation and hemoglobin concentration measurements in Gum 1 and Gum 3 chewing. One example of Gum 1 chewing appears to show no changes in oxygen saturation levels as well as hemoglobin concentration during chewing. The other example of Gum 3 chewing showed that oxygen saturation level rapidly decreases from the start of chewing, then becomes stable with a break point between the two states, followed by an increase after the end of chewing. Total hemoglobin and oxyhemoglobin concentrations increase during chewing, while deoxyhemoglobin level increases up to the break point and then becomes constant and decreases after finishing chewing.

Changes in muscle oxygen saturation in the 23 subjects are summarized in Table I. For the chewing of Gum 1, none of the subjects showed any significant differences in oxygen saturation between 10 s before chewing and 10 s before finishing

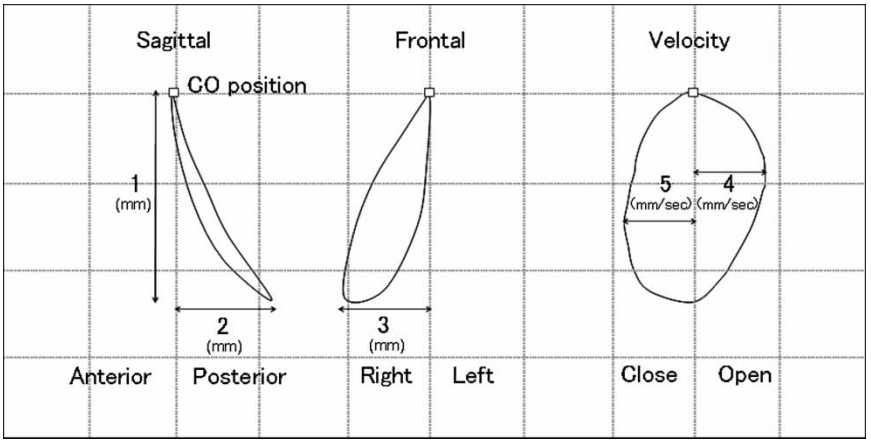

Figure 2. Analysis of mandibular movement using the mandibular kinesiograph. 1. Maximum vertical opening from CO. 2. Maximum antero-posterior movement from CO. 3. Maximum lateral deviation. 4. Maximum opening velocity. 5. Maximum closing velocity.

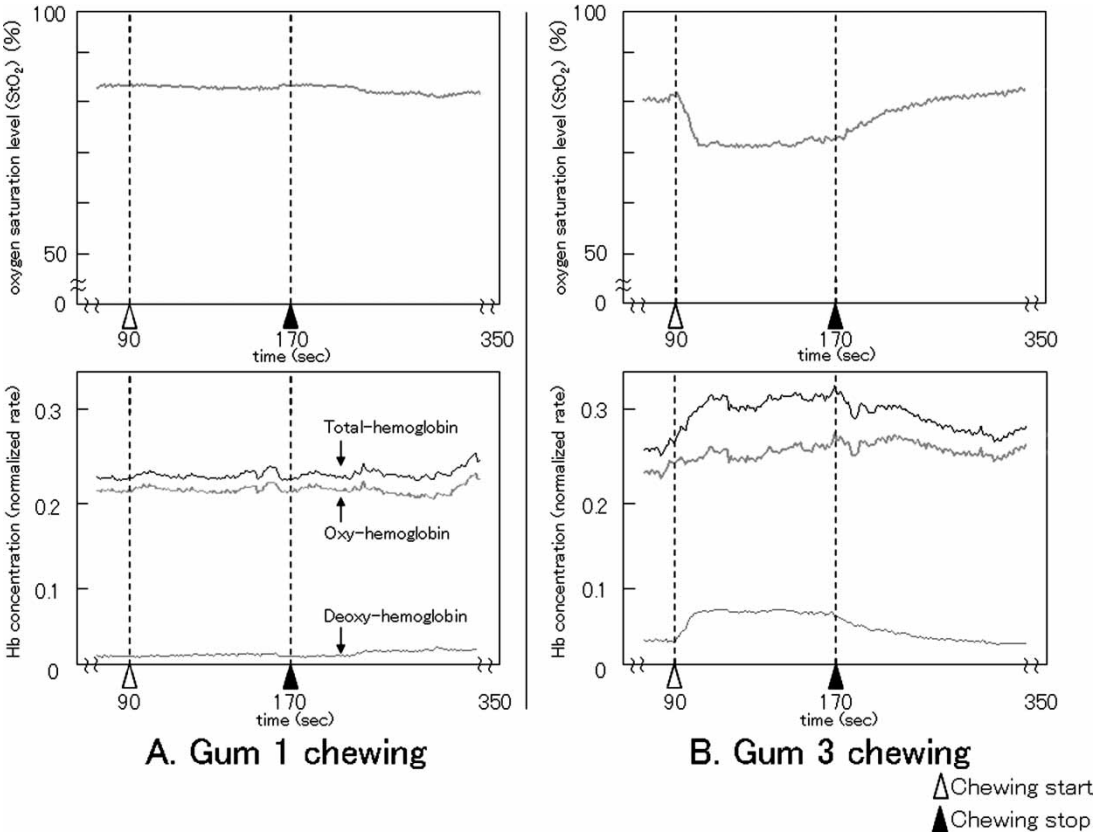

Figure 3. Typical examples of muscle oxygenation (upper graph) and hemoglobin concentration (lower graph) changes while chewing Gum 1 (A) and Gum 3 (B). Examples A and B were obtained from measurements in different subjects.

Table I. Changes in muscle oxygen saturation in the 23 subjects.

	No change	Decreasing
Gum 1	23	0
Gum 2	10*	13*
Gum 3	0	23

*Subjects were divided into two groups according to masseter muscle oxygen saturation changes while chewing Gum 2: Group 1 ($n=10$), no significant changes; and Group 2 ($n=13$), significant decreases.

chewing. For the chewing of Gum 2, 10 subjects showed no significant differences (Group 1), whereas the other 13 subjects showed significant decreases in oxygen saturation levels during gum chewing (Group 2). For the chewing of Gum 3, significant decreases were revealed in all subjects. In all cases with decreasing oxygen saturation, the break point at which oxygen saturation level stops decreasing and becomes constant, or desaturation slows, was found. By drawing two tangents drawn from the start and finish of chewing on the oxygen saturation curve, the time of break point appearance could be identified as an intersection of the two tangents (Figure 4). Break point appearance time ranged from 18.0 s to 28.0 s for Gum 3 chewing in all 23 subjects, and from 19.0 s to 29.0 s for Gum 2 chewing in the 13 subjects of Group 2.

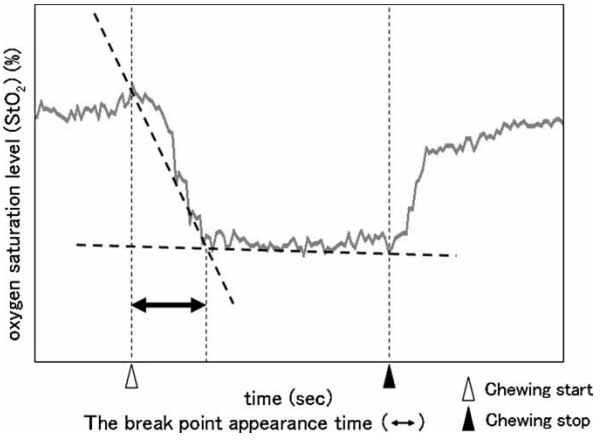

Figure 4. Calculation of the break point appearance time. By drawing two tangents from the start and finish of chewing on the oxygen saturation curve, break point appearance time was identified as an intersection of the two tangents.

Mandibular movement during gum chewing

The five mandibular movement parameters in each chewing were determined as mean values at an interval of 30 strokes. To investigate relationships between muscle oxygen saturation state and mandibular movement, 30-stroke intervals were set so as to be relatable to muscle oxygen states in the three kinds of gum chewing in each of the two groups. For Gum 3 chewing, the interval was set so that the first stroke was the one just after appearance of the break

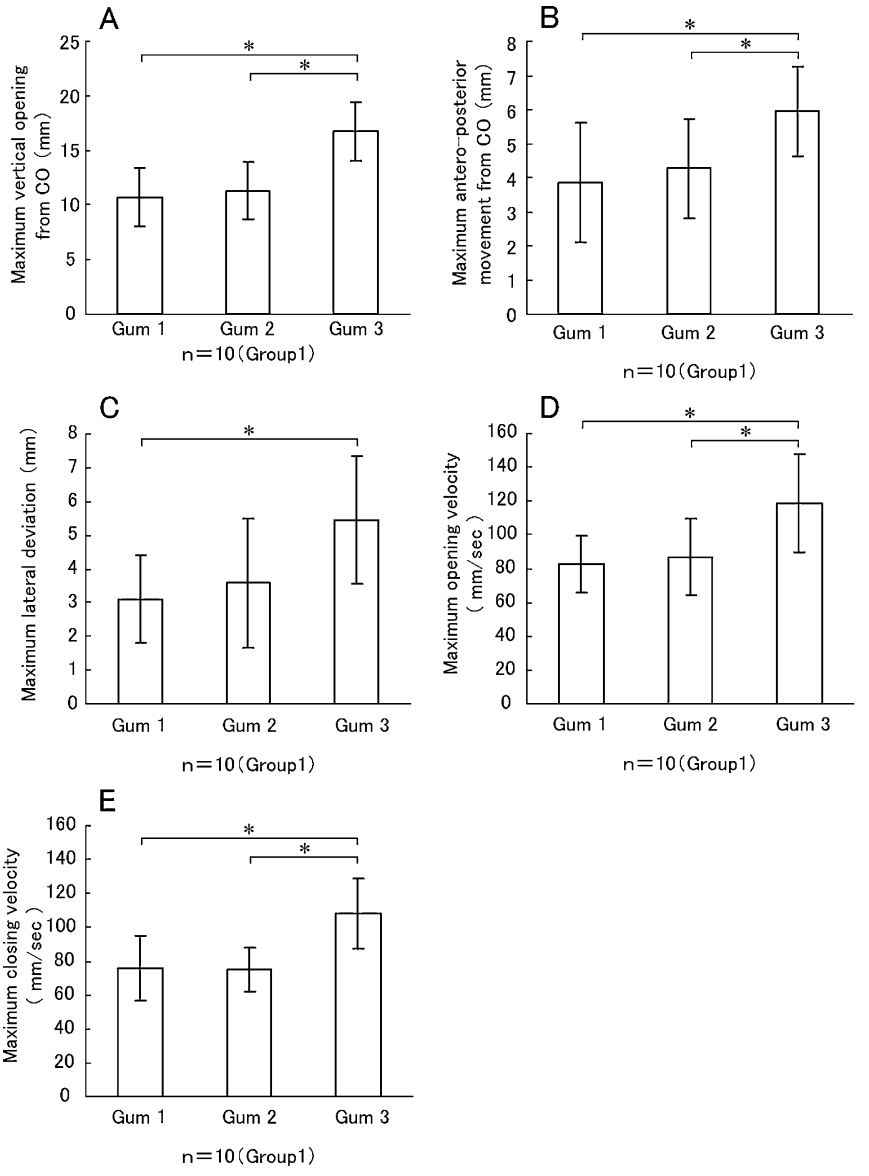

Figure 5. Mean values and SD of maximum vertical opening from CO (A), maximum antero-posterior movement from CO (B), maximum lateral deviation (C), maximum opening velocity (D), and maximum closing velocity (E) for the chewing of Gum 1, Gum 2, and Gum 3 in Group 1 with level of significance ($*p < 0.05$).

point. For Gum 1 chewing, the 30 strokes corresponding to Gum 3 chewing was used. For Gum 2 chewing, in Group 1 the 30 strokes corresponding to Gum 3 chewing was used, whereas in Group 2 the interval was set just after the appearance of the break point.

Figures 5 and 6 show comparisons of mandibular movement parameters among the chewing of Gums 1, 2, and 3 in Groups 1 and 2, respectively. In Group 1, all 5 parameters were significantly larger in Gum 3 chewing than in Gum 1 chewing. Except for maximum lateral deviation, 4 parameters showed significantly larger values in Gum 3 chewing than in Gum 2 chewing. No significant differences in any parameters were seen between Gum 1 and Gum 2 chewing. In Group 2, both Gum 2 and Gum 3 chewing showed significantly larger values in all parameters than Gum 1 chewing. No significant differences in any parameters were seen between Gum 2 and Gum 3 chewing.

Discussion

In this experiment, gum was used as a test food because it enables continuous chewing for measuring masseter muscle oxygen saturation and mandibular movement. Prior to the experiment, masseter muscle oxygen saturation changes were monitored in 8 randomly selected subjects using the same method described above, using 4 kinds of gum: Gum 1; Gum 2; Gum 3; and a mix of 50% Gum 1 and 50% Gum 3. For Gum 1 chewing, all 8 subjects showed no apparent changes in oxygen saturation levels. For Gum 2 chewing, some subjects showed no apparent changes, but others showed decreased oxygen saturation. In 50% mixed gum chewing, all subjects

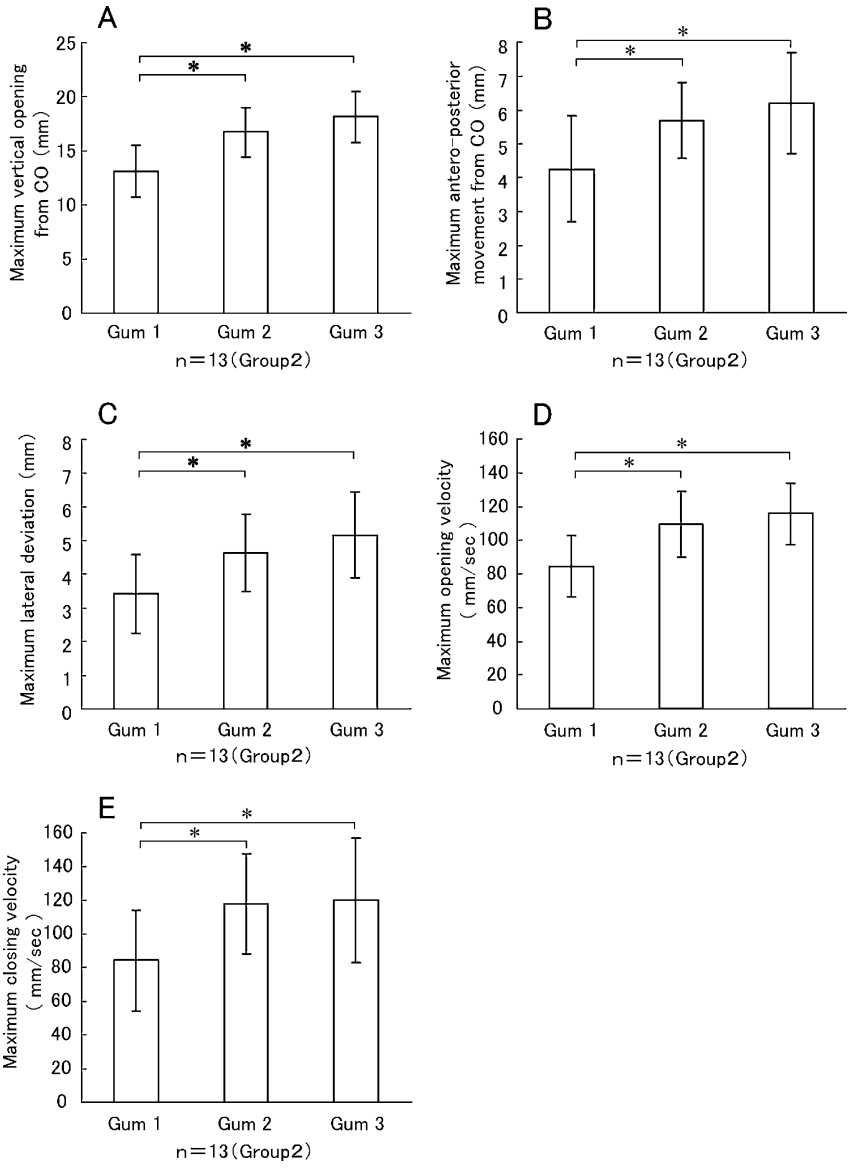

Figure 6. Mean values and SD of maximum vertical opening from CO (A), maximum antero-posterior movement from CO (B), maximum lateral deviation (C), maximum opening velocity (D), and maximum closing velocity (E) for the chewing of Gum 1, Gum 2, and Gum 3 in Group 2 with level of significance ($^{*}p < 0.05$).

showed decreased oxygen saturation as in Gum 3 chewing. Expecting different effects of gum hardness on muscle oxygenation, this experiment employed Gums 1, 2 and 3 as test materials. Regarding the reliability of NIRS measurements of masseter muscle oxygenation, the previous study [26] evaluated method errors for repeated measurements of break point appearance time during clenching, and confirmed reproducibility of the methods.

Skeletal muscle fibers are classified as slow-twitch (type 1) and fast-twitch (type 2) according to the rate of contraction [34,35]. Slow-twitch fibers have a dense capillary network and are rich in both mitochondria and myoglobin. Myoglobin aids in the transfer of oxygen from hemoglobin in blood to the mitochondria in muscle cells, thus facilitating ATP generation through oxidative (aerobic) metabolism. Type 1 fibers mostly rely on oxidative metabolism for their energy supply and are resistant to fatigue. Fast-twitch fibers are further subclassified into types 2A and 2B. Type 2A fibers are rich in mitochondria and myoglobin and have a good blood supply, as well as high myosin ATPase activity. Type 2A fibers rely on oxidative metabolism for energy and are relatively resistant to fatigue. Type 2B fibers have a high myosin ATPase activity, large quantities of glycogen, and high concentrations of glycolytic enzymes, but fewer mitochondria and lower concentrations of myoglobin, and a limited blood supply. Type 2B fibers mostly rely on glycolytic (anaerobic) metabolism for energy. This anaerobic metabolism is accomplished by the accumulation of lactate and thus a decrease of intracellular pH. This lactic acidosis impedes force generation [34]. The by-products of glycolytic metabolism are considered responsible for the progressive impairment of the

muscle contractile process, so type 2B fibers are easily fatigued. Most skeletal muscles are composed of fibers of each twitch type. Several studies [36–38] have reported fiber-type compositions of human masseter muscle. Eriksson & Thornell [37] showed a predominance of type 1 fibers in the masseter muscle with variations in fiber composition among different parts of the muscle and among subjects.

In work and exercise, the proportion of aerobic and anaerobic metabolism and the consumption of oxygen for energy supply vary with the severity and duration of the work done [35]. Masseter muscle oxygen saturation changes relate to energy metabolism and oxygen consumption in gum chewing with different hardness. For the chewing of Gum 1, no subjects showed any significant changes in oxygen saturation. As Gum 1 is a chewing gum with lower hardness, smaller force seems sufficient for chewing. As a pattern of force generation, the size principle [34] explains that the brain first recruits smaller motor units that consist of slow-twitch (type 1) fibers for less force generation, then larger motor units that contain fast-twitch (type 2) fibers are recruited and force generation increases accordingly. Accordingly, chewing force would be generated mainly by slow-twitch (type 1) fibers during Gum 1 chewing, and oxygen consumption for aerobic metabolism would be low because of the lower force needed. The oxygen saturation curve during Gum 1 chewing thus seems to indicate a balanced state between oxygen supply and consumption in the muscle.

For the chewing of Gum 3, significant decreases in muscle oxygen saturation were shown in all subjects. Chewing force would become greater with increased hardness of the gum, and thus should be developed by both slow- and fast-twitch fibers. Muscle oxygen saturation levels rapidly decreased from the start of chewing, which seems to indicate that oxygen consumption for aerobic metabolism proceeded rapidly. Thereafter, desaturation stopped or slowed, showing a break point between the two phases. Total hemoglobin concentrations increased throughout chewing, probably because strong rhythmic contractions increased blood flow through the muscle [35]. Deoxygenated hemoglobin level increased up to the break point and then became almost constant, indicating that changes of oxygenated hemoglobin to deoxygenated hemoglobin for oxygen supply became reduced at the break point. This suggests that fast-twitch glycolytic (type 2B) fibers play a major role in sustaining chewing after the break point. For Gum 2 chewing, individual variations were seen in masseter muscle oxygen saturation changes. Ten subjects (Group 1) showed no apparent changes, as in Gum 1 chewing, while the remaining subjects (Group 2) showed decreases in oxygen saturation, as in Gum 3 chewing. In Group 1, sufficient force would be generated by slow-twitch fibers for the chewing of Gum 2, whereas in Group 2 slow-twitch fibers alone might be unable to generate sufficient force and tension of fast-twitch fibers would be needed. The difference in muscle oxygen saturation changes between Groups 1 and 2 may be due to differences in oxidative capacity of masseter muscle, such as the number of mitochondria and the amount of myoglobin in the muscle and capillary density surrounding the muscle, and in the number of myofibrils in individual slow-twitch fibers as well as fiber composition [39].

Mandibular movement parameters were compared in different situations of masseter muscle oxygen saturation changes during gum chewing in both Groups 1 and 2. Attempting to compare mandibular movements developed by different kinds of metabolism for energy supply, intervals of chewing strokes to be analyzed were set in reference to a break point on the oxygen saturation curve. Studies have reported that harder food enlarges chewing motion [1–5], increases velocity of chewing motion [3,6,7], and increases electromyographic activity of the masseter muscle [2,4,8,9]. In this study, both Groups 1 and 2 showed that chewing motions were larger and velocity higher in mandibular movement, probably resulting from an increase in the contribution of fast-twitch fibers with anaerobic metabolism to energy supply. Fast-twitch glycolytic (type 2B) fibers develop great tension rapidly, but fatigue also sets in rapidly, over the order of tens of seconds or minutes, reflecting the inability of fibers to maintain tension owing to rapid declines in glycogen and decreased local pH [34]. Masseter muscle fatigue readily occurs during gum chewing, where muscle force is generated by fast-twitch glycolytic fibers (type 2B) in relation to gum hardness.

The chewing motion in this study is experimental rather than physiological because of the regulation of chewing rhythm, duration of chewing, and chewing side. However, in measurements of masseter muscle oxygen saturation and mandibular movement, the results suggest that hardness of the test bolus influences the pattern of oxygen consumption and energy metabolism in masseter muscle, resulting in changing amplitude of chewing motion and chewing velocity. Variations exist in these responses to bolus hardness, which may indicate individual differences in muscle fatigue tendency for harder food chewing.

Further studies will be conducted to investigate the influences of other food textures, such as size, shape, cohesiveness, gumminess, flexibility, and adhesiveness, on masticatory muscle oxygen saturation for a better understanding of muscle energy metabolism in mastication. Furthermore, NIRS measurement of masticatory muscle oxygenation during chewing of some test boluses could be expected to evaluate individual fatigue tendencies of masticatory muscles.

Conclusions

The results suggest that the harder texture of gum enlarges chewing motion and increases chewing velocity with an increase in the contribution of anaerobic metabolism to energy yield in masseter muscle. Differences in the responses to gum hardness may indicate individual differences in muscle fatigue tendencies when chewing harder foods.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

References

[1] Gibbs CH, Wickwire NA, Jacobson AP, Lundeen HC, Mahan PE, Lupkiewicz SM. Comparison of typical chewing patterns in normal children and adults. J Am Dent Assoc 1982;105:33–42.

[2] Horio T, Kawamura Y. Effects of texture of food on chewing patterns in the human subject. J Oral Rehabil 1989;16: 177–83.

[3] Anderson K, Throgkmorton GS, Buschang PH, Hayasaky H. The effects of bolus hardness on masticatory kinematics. J Oral Rehabil 2002;29:689–96.

[4] Peyron MA, Blanc O, Lund JP, Woda A. Influence of age on adapatability of human mastication. J Neurophysiol 2004;92: 773–9.

[5] Häggman-Henrikson B, Eriksson PO. Head movements during chewing: relation to size and texture of bolus. J Dent Res 2004;83:864–8.

[6] Chew CL, Lucas PW, Tay DK, Keng SB, Ow RK. The effect of food texture on the replication of jaw movements in mastication. J Dent 1988;16:210–4.

[7] Peyron MA, Maskawi K, Woda A, Tanguay R, Lund JP. Effects of food texture and sample thickness on mandibular movement and hardness assessment during biting in man. J Dent Res 1997;76:789–95.

[8] Plesh O, Bishop B, McCall W. Effect of gum hardness on chewing pattern. Exp Neurol 1986;92:502–12.

[9] Ottenhoff FA, Van Der Bilt A, Van Der Glas HW, Bosman F. Control of elevator muscle activity during simulated chewing with varying food resistance in humans. J Neurophysiol 1992;68:933–44.

[10] Barcroft H, Millen JL. The blood flow through muscle during sustained contraction. J Physiol 1939;97:17–31.

[11] Byström SE, Kilbom A. Physiological response in the forearm during and after isometric intermittent handgrip. Eur J Appl Physiol Occup Physiol 1990;60:457–66.

[12] Hogan MC, Richardson RS, Kurdak SS. Initial fall in skeletal muscle force development during ischemia is related to oxygen availability. J Appl Physiol 1994;77:2380–4.

[13] Mancini DM, Bolinger L, Li H, Kendrick K, Chance B, Wilson JR. Validation of near-infrared spectroscopy in humans. J Appl Physiol 1994;77:2740–7.

[14] Belardinelli R, Barstow TJ, Porszasz J, Wasserman K. Skeletal muscle oxygenation during constant work rate exercise. Med Sci Sports Exerc 1995;27:512–9.

[15] Belardinelli R, Barstow TJ, Porszasz J, Wasserman K. Changes in skeletal muscle oxygenation during incremental exercise measured with near infrared spectroscopy. Eur J Appl Physiol Occup Physiol 1995;70:487–92.

[16] Maehara K, Riley M, Galassetti P, Barstow TJ, Wasserman K. Effect of hypoxia and carbon monoxide on muscle oxygenation during exercise. Am J Respir Crit Care Med 1997;155:229–35.

[17] Rundell KW, Nioka S, Chance B. Hemoglobin/myoglobin desaturation during speed skating. Med Sci Sports Exerc 1997;29:248–58.

[18] Bhambhani YN, Buckley SM, Susaki T. Detection of ventilatory threshold using near infrared spectroscopy in men and women. Med Sci Sports Exerc 1997;29:402–9.

[19] Grassi B, Quaresima V, Marconi C, Ferrari M, Cerretelli P. Blood lactate accumulation and muscle deoxygenation during incremental exercise. J Appl Physiol 1999;87:348–55.

[20] Bhambhani Y, Maikala R, Esmail S. Oxygenation trends in vastus lateralis muscle during incremental and intense anaerobic cycle exercise in young men and women. Eur J Appl Physiol 2001;84:547–56.

[21] Fadel PJ, Keller DM, Watanabe H, Raven PB, Thomas GD. Noninvasive assessment of sympathetic vasoconstriction in human and rodent skeletal muscle using near-infrared spectroscopy and Doppler ultrasound. J Appl Physiol 2004;96:1323–30.

[22] Lorenz KA, Hom C, Lopez J, Pozos RS, Buono MJ, Kolkhorst FW. Effects of hypoxia on the onset of muscle deoxygenation and the lactate threshold. J Physiol Sci 2006; 56:321–3.

[23] Chance B, Cope M, Gratton E, Ramanujim N, Tromberg B. Phase measurement of light absorption and scatter in human tissue. Rev Sci Instrum 1998;69:3457–81.

[24] Delcanho RE, Kim YJ, Clark GT. Haemodynamic changes induced by submaximal isometric contraction in painful and non-painful human masseter using near-infra-red spectroscopy. Arch Oral Biol 1996;41:585–96.

[25] Sugisaki M, Misawa A, Ikai A, Young-Sung K, Tanabe H. Sex differences in the hemoglobin oxygenation state of the resting healthy human masseter muscle. J Orofac Pain 2001; 15:320–8.

[26] Yanagihara K, Ishikawa H, Toyama M. Comparison of masseter muscle oxygenation during clenching between normal occlusion and open bite subjects using near-infrared spectroscopy. Orthodontic Waves 2004;63:101–6.

[27] Tsukiyama Y, Kuboki T, Clark GT. Cold pressor stimulation effect on hemodynamic changes following sustained isometric contraction in human jaw-closure muscles. J Dent Res 1999;78:1727–34.

[28] Inoue-Minakuchi M, Maekawa K, Kuboki T, et al. Intramuscular haemodynamic responses to different durations of sustained extension in normal human masseter. Arch Oral Biol 2001;46:661–6.

[29] Kuboki T, Suzuki K, Maekawa K, et al. Correlation of the near-infrared spectroscopy signals with signal intensity in T(2)-weighted magnetic resonance imaging of the human masseter muscle. Arch Oral Biol 2001;46:721–7.

[30] Aizawa S, Tsukiyama Y, Koyano K, Clark GT. Reperfusion response changes induced by repeated, sustained contractions in normal human masseter muscle. Arch Oral Biol 2002;47:537–43.

[31] Hidaka O, Yanagi M, Takada K. Changes in masseteric hemodynamics time-related to mental stress. J Dent Res 2004;83:185–90.

[32] Okada K, Yamaguchi T, Komatsu K, Matsuki T, Gotouda A, Minowa K, Inoue N. The influence of tissue blood flow volume on energy metabolism in masseter muscles. Cranio 2005;23:166–73.

[33] Lavelle CL. Applied oral physiology, 2nd edn. London: Wright; 1988. p. 12–24.

[34] Boron WF, Boulpaep EL. Medical physiology. A cellular and molecular approach. Philadelphia: Saunders; 2003. p. 250–3. p. 1242–7.

[35] Pocock G, Richards CD, Daly MB. Human physiology. The basis of medicine, 2nd edn. New York: Oxford Univ Press; 2004. p. 102–7, p. 579–83.

[36] Ringqvist M, Ringqvist I, Eriksson PO, Thornell LE. Histochemical fibre-type profile in the human masseter muscle. J Neurol Sci 1982;53:273–82.

[37] Eriksson PO, Thornell LE. Histochemical and morphological muscle-fibre characteristics of the human masseter, the medial pterygoid and the temporal muscles. Arch Oral Biol 1983;28:781–95.

[38] Stålberg E, Eriksson PO, Antoni L, Thornell LE. Electrophysiological study of size and fibre distribution of motor units in the human masseter and temporal muscles. Arch Oral Biol 1986;31:521–7.

[39] Johnson LR. Essential medical physiology, 3rd edn. San Diego, CA: Elsevier Academic Press; 2003. p. 939–40.