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# ORIGINAL

# Haemodynamic reactions in human masseter muscle during different types of contractions

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#### ABSTRACT :

Objectives : To investigate to what extent different types of jaw-muscle contractions cause haemodynamic reactions in human masseter muscle. Materials and Methods : Eleven healthy volunteers (seven males : 25.0 ± 2.9 years and four females: 23.3 ± 4.3 years) performed three standardized oral-motor tasks : maximal voluntary contractions (MVC ; duration 5 sec, 3 times repetition), tooth grinding (repetitive left and right side grinding from intercuspal position to canine-to-canine position at 0.5 Hz keeping 50% MVC for a total of 10 times), and 1-min left-side gum chewing at 1 Hz. Haemodynamic characteristics were measured in the left masseter muscle with the use of a laser blood oxygenation monitor (BOM-L1TRW, OMEGAWAVE INC., Tokyo, Japan). Electromyographic (EMG) activity from right and left masseter muscle was simultaneously monitored (500 Hz sample frequency) during the tasks. 1-ANOVA followed by Dunnett's test was used. Results : Oxygenated haemoglobin (OXYHb : 13.5 ± 0.2 10<sup>4</sup> units/mm<sup>3</sup>) and deoxygenated haemoglobin (deOXYHb : 7.6  $\pm$  0.3 10<sup>4</sup> units/mm<sup>3</sup>) did not change significantly during the MVC task (13.9  $\pm$  0.2 and 7.8  $\pm$  0.3 10<sup>4</sup> units/mm<sup>3</sup>, respectively, P>0.065), however, the total haemoglobin (TOTALHb : 22.1 ± 0.3 10<sup>4</sup> units/mm<sup>3</sup>) showed a significant increase (22.7±0.3 10<sup>4</sup> units/mm<sup>3</sup>, P=0.003) during the MVC. Tissue blood oxygen saturation was not changed during the MVC (P=0.164). During the tooth grinding task, OXYHb, deOXYHb, TOTALHb, and tissue blood oxygen saturation (StO<sub>2</sub>) remained constant (P>0.127). Finally, the chewing task was associated with significant decreases in StO<sub>2</sub> (67.9±0.7%, P=0.006) related to a decrease in OXYHb ( $14.0 \pm 0.2 \ 10^4$  units/mm<sup>3</sup>, P=0.040) compared to baseline ( $68.8 \pm 0.7\%$  and 14.2 $\pm 0.3 \ 10^4$  units/mm<sup>3</sup>, respectively). Conclusion : These results showed that high-intensity experimental tooth clenching caused constriction-like reactions in the masseter muscle whereas tooth grinding did not cause detectable changes in haemodynamic characteristics of masseter muscle. Finally, the findings indicated that rhythmic dynamic contractions might lead to oxygen deficit in the masseter muscle. The present data may have implications for understanding the potential pathophysiological consequences of different types of oral-motor tasks, e.g., bruxism and prolonged mastication.

Key Words : blood oxygenation, haemodynamic characteristics, maximal voluntary contraction, masseter muscle.

# INTRODUCTION

Pain and/or soreness in masticatory muscle are significant symptoms and signs in patients with functional disorders of the chewing apparatus, e.g. myofascial pain and dysfunction, which recently has been coined persistent orofacial muscle pain (POMP)<sup>1)</sup>. The main cause of POMP has often been thought to be the overuse of jaw muscles during parafunctional activity, such as tooth grinding and clenching, i.e., bruxism<sup>2, 3)</sup>. It is generally considered that vasodilatation and vasoconstriction are basic physiological adjustments to the metabolic demands

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of skeletal muscles<sup>4)</sup> and ischemic pain occurs when there is insufficient blood flow for the metabolic needs of the muscle<sup>5-7)</sup>. This theory has also been adapted to the jawclosing muscles as a prototypical example<sup>8-10)</sup>. In fact, there are some reports that individuals with a history of chronic jaw-muscle pain linked to dysfunction of the sympathetic nervous system have slow intramuscular reperfusion during the recovery phase after sustained isometric contractions<sup>9, 11)</sup>. Multiple neuroactive compounds such as muscle phosphorylase, debrancher enzyme, phosphofructokinase, phosphoglycerate kinase, and lactate dehydrogenase can be released from an ischemic muscle and could contribute to pain by activations of nociceptive afferents in the muscle tissue<sup>4)</sup> but it has been questioned whether these changes are sufficient to trigger pain.

In a previous study, intramuscular blood flow during different levels of masseter muscle contractions were measured and the results showed that masticatory muscle haemodynamics during different levels and types of contraction lead to slight increases of blood flow<sup>12</sup>). This haemodynamic increase was also observed in other studies<sup>13-16</sup>. Nevertheless, the effect of different types of muscle contractions on blood flow in jaw-closing muscle is still controversial.

The aim of the present study was to measure the haemodynamic reactions during maximal voluntary contractions based on the hypothesis that blood flow during a sustained and high-intensity isometric contraction would be insufficient to meet the metabolic demand, resulting in relative local ischaemia with the development of a strong reflex hyperaemia after the contraction. Furthermore, it was presumed that these hemodynamic responses would be dependent on the specific type of jaw-muscle contractions so in addition to MVC, the consequences of tooth-grinding and rhythmic mastication were also tested.

# MATERIALS AND METHODS

## Subjects

Eleven healthy volunteers aged 19-28 years (seven males : mean  $\pm$  SD, 25.0  $\pm$  2.9 years old and four females : 23.3  $\pm$  4.3 years old) participated in this study. All were from a university population and were in good general health. The absence of dental disease and systemic disorders were ascertained by questionnaires and clinical examination and the Research Diagnostic Criteria for Temporomandibular Disorders (RDC/TMD) were used to verify the absence of painful conditions in the jaw muscles and temporomandibular joint<sup>17)</sup>. Furthermore, it was ascertained that none of the subjects took medication which could affect pain or vascular responses during the experimental period. Informed consent was obtained from each subject and the experimental protocol was approved by local ethics committee.

#### Experimental protocol

The subjects sat on a comfortable chair in a sound attenuated room, with controlled temperature and performed three standardized oral-motor tasks: maximal voluntary contractions (MVC), tooth grinding, and gum chewing. Each task was separated by at least 5-min rest. Bilateral masseter electromyographic (EMG) activity and blood oxygenation (BO) levels from the left masseter muscle were simultaneously recorded. The changes of BO during the oral-motor tasks were off-line analysed and compared to that from baseline and immediately after the tasks (recovery).

## Oral-motor tasks

#### Maximal voluntary contractions

The subjects were asked to clench their teeth as hard as possible for 5 sec and to repeat the MVC for three times. Verbal encouragement was performed in order to obtain the maximal effort<sup>18)</sup>. Each contraction was followed by rest for about 1 sec.

#### Tooth grinding

The tooth-grinding task was repetitive tooth grinding from the intercuspal position to the left side canine-tocanine position at 0.5 Hz with approximately 50% MVC for a total of 10 times. The pace of the tooth grinding was guided by sounds from a metronome. The voltage of approximately 50% MVC was visually feedback through a pc monitor (target level and actual masseter muscle activity (EMG)). After 2 min rest, tooth grinding was shifted to the right side canine-to-canine position and repeated at 0.5 Hz with 50% MVC for a total of 10 times.

## Gum chewing

The mastication task was chewing a gum (Green Gum, Lotte, Tokyo, Japan) only on the left side. After an initial adaptation period of approximately 2 min to account for the initial change in softness of the chewing gum, subjects started the chewing task and continued for 1 min at a frequency of 1 Hz guided by a metronome. In this task, the baseline recording was performed before the 2-min of initial adaptation.

## Bilateral masseter EMG activity

The EMG activity in the superficial parts of the left and right masseter muscles were recorded with surface EMG electrodes. The skin was cleaned with ethanol and bipolar disposable surface electrodes (F-150F, NIHON KOHDEN, Tokyo, Japan) were placed with their long axis paralleltransverse to the main fibre direction of the right and left masseter muscles (Fig. 1) based on palpation of the muscles during a full effort of muscle contraction<sup>19</sup>. The EMG signals were amplified (6.4 x  $10^6$  times) and filtered (0.53-250 Hz) and then were simultaneously recorded during the experiments through analogue-todigital convertor with a sample frequency (500 Hz) by a processor box (5201, NF Corporation, Yokohama, Japan) and stored in a PC.

# Blood oxygenation

Haemodynamic characteristics were measured from the left masseter muscle with the use of a laser blood oxygenation monitor (BOM-L1TRW, OMEGAWAVE, Tokyo, Japan, Fig. 1). The BO recording device has one probe and two detectors. The distance between the probe to the nearest detector was 1 cm, and 2 cm to the second detector so that BO between the depth of 1 cm and 2 cm from the skin surface can be measured  $^{20)}$ . After collecting the signals from these two layers (between 0 cm to 1 cm depth from the skin surface and between 0 cm to 2 cm depth from the skin surface), the targeted data (between 1 cm to 2 cm depth from the skin surface = the placement of masseter muscle fibre) was manually calculated using a special formula provided by the company (OMEGAWAVE, Tokyo, Japan). The following parameters can be derived from the BO monitoring

Locations of electrodes for EMG and probes and detectors for BO

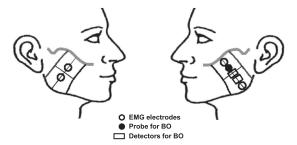


Fig. 1 Schematic set-up of experimental measures. EMG : electromyographic (activity). BO : blood oxygenation (recording).

device: oxygenated haemoglobin (OXYHb), deoxygenated haemoglobin (deOXYHb), total haemoglobin (TOTALHb), and tissue blood oxygen saturation (StO<sub>2</sub>).

## Statistical analysis

The haemodynamic characteristics were evaluated by one-way analysis of variance (1-ANOVAs) with repeated measures followed by Dunnett's test. The variable was "time" and 10 sec before each oral-motor task was defined as the baseline level (Baseline), whole time of each oralmotor task was defined as experimental level (During), and 10 sec after each oral-motor task was defined as the post period (Recovery) and used for the post-hoc tests. Parametric statistics (mean  $\pm$  standard deviation (S.D.)) were used to describe the data and the level of significance was set to P<0.05.

## RESULTS

## Effect of maximal voluntary contractions

Fig. 2 shows the raw signals of EMG and BO during MVC task from one subject (Subject No. 11). OXYHb and deOXYHb (Baseline :  $13.5 \pm 2.0 \ 10^4 \ \text{units/mm}^3$  and  $7.6 \pm 2.5 \ 10^4 \ \text{units/mm}^3$ ) did not change significantly during MVC ( $13.9 \pm 2.1 \ 10^4 \ \text{units/mm}^3$  and  $7.8 \pm 2.7 \ 10^4 \ \text{units/mm}^3$ , 1-ANOVA P>0.065, Fig. 3-C and -D). However,

#### EMG and BO signals during maximal voluntary contraction

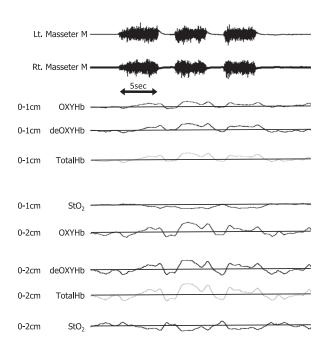


Fig. 2 Electromyographic (EMG) activity and changes of blood oxygenation (BO) during maximal voluntary contraction from one subject (Subject No. 11).

TOTALHb (Baseline :  $22.1 \pm 3.5 \ 10^4$  units/mm<sup>3</sup>) showed a significant increase during the MVC ( $22.7 \pm 3.4 \ 10^4$  units/mm<sup>3</sup>, 1-ANOVA P=0.003, Fig. 3-B) and decreased to baseline level during Recovery ( $22.3 \pm 3.2 \ 10^4$  units/mm<sup>3</sup>). Finally, StO<sub>2</sub> (Baseline :  $66.6 \pm 8.1\%$ ) did not change during the MVC task ( $66.8 \pm 8.7\%$ ) or post task period (Recovery :  $68.0 \pm 9.4\%$ , 1-ANOVA P=0.164, Fig. 3-A).

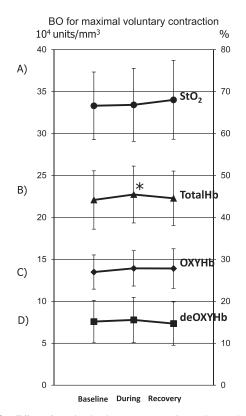
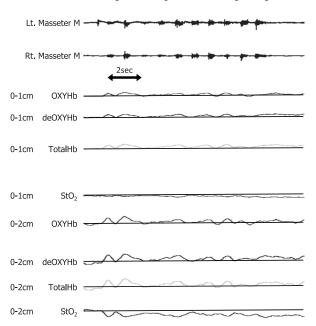


Fig. 3 Effect of maximal voluntary contraction on haemodynamic in left masseter muscle (Mean±S.D.). A. Changes in StO<sub>2</sub> (tissue blood oxygen saturation, OXYHb(oxygenated haemoglobin)/TOTALHb(total haemoglobin)×100). B. Changes in TOTALHb. C. Changes in deOXYHb (deoxygenated haemoglobin). D. Changes in OXYHb. Dunnett's test : \* P<0.050.</p>

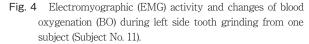
# Tooth grinding

Fig. 4 shows the raw signals of EMG and BO during the tooth-grinding task (subject No. 11).

During the left side tooth-grinding task, the ipsilateral (left side) masseter OXYHb (Baseline :  $13.8 \pm 1.8 \ 10^4$  units/mm<sup>3</sup>) did not change ( $13.7 \pm 1.9 \ 10^4$  units/mm<sup>3</sup>, 1-ANOVA P=0.127, Fig. 5-C). The deOXYHb (Baseline :  $7.6 \pm 2.6 \ 10^4$  units/mm<sup>3</sup>) did not change during the task ( $7.5 \pm 2.6 \ 10^4$  units/mm<sup>3</sup>) or the recovery phase (Recovery :  $7.6 \pm 2.6 \ 10^4$  units/mm<sup>3</sup>, 1-ANOVA P=0.408, Fig. 5-D). TOTALHb (Baseline :  $22.4 \pm 3.3 \ 10^4$  units/mm<sup>3</sup>) did not change during the task ( $22.3 \pm 3.3 \ 10^4$  units/mm<sup>3</sup>, 1-ANOVA P=0.136,



EMG and BO signals during left side tooth grinding



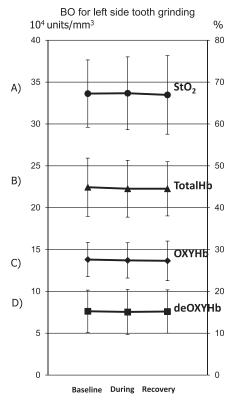


Fig. 5 Effect of left side tooth grinding on haemodynamic in left masseter muscle (Mean ± S.D.). A. Changes in StO<sub>2</sub> (tissue blood oxygen saturation, OXYHb(oxygenated haemoglobin)/TOTALHb(total haemoglobin)×100). B. Changes in TOTALHb. C. Changes in deOXYHb (deoxygenated haemoglobin). D. Changes in OXYHb.

Fig. 5-B). StO<sub>2</sub> (Baseline :  $67.3 \pm 8.6\%$ ) also did not change during tooth grinding ( $67.3 \pm 8.2\%$ ) or post task period (Recovery :  $66.9 \pm 8.7\%$ , 1-ANOVA P=0.277, Fig. 5-A).

For the right side tooth grinding, contralateral (left side) masseter OXYHb, deOXYHb, TOTALHb, or  $StO_2$  (Baseline :  $13.7 \pm 2.2 \ 10^4$  units/mm<sup>3</sup>,  $7.4 \pm 2.5 \ 10^4$  units/mm<sup>3</sup>,  $22.1 \pm 3.2 \ 10^4$  units/mm<sup>3</sup>,  $67.6 \pm 9.0\%$ , respectively) were not changed by the task ( $13.9 \pm 2.0 \ 10^4$  units/mm<sup>3</sup>,  $7.4 \pm 2.5 \ 10^4$  units/mm<sup>3</sup>,  $22.3 \pm 3.3 \ 10^4$  units/mm<sup>3</sup>,  $68.1 \pm 8.4\%$ , respectively, 1-ANOVA P>0.262).

## Gum chewing

Fig. 6 shows the raw signals of EMG and BO during the gum chewing task (subject No. 11). OXYHb (Baseline : 14.2  $\pm 2.6 \ 10^4$  units/mm<sup>3</sup>) was not affected by the task (14.0  $\pm 2.4 \ 10^4$  units/mm<sup>3</sup>, Fig. 7-C), however, significantly decreased during the post task period (Recovery :  $13.9 \pm 2.4 \ 10^4$  units/mm<sup>3</sup>, 1-ANOVA P=0.040). The deOXYHb and TOTALHb (Baseline :  $7.3 \pm 2.3 \ 10^4$  units/mm<sup>3</sup> and  $22.5 \pm 3.7 \ 10^4$  units/mm<sup>3</sup>, respectively, Fig. 7-D and -B). However, the task was associated with significant decreases in StO<sub>2</sub> (Baseline :  $68.8 \pm 7.3\%$ ) during the task (67.9  $\pm 7.2\%$ ) and recovery period (Recovery :  $67.6 \pm 7.6\%$ , 1-ANOVA P=0.006, Fig. 7-A).

#### EMG and BO signals during gum chewing

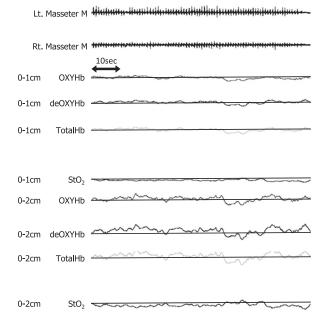
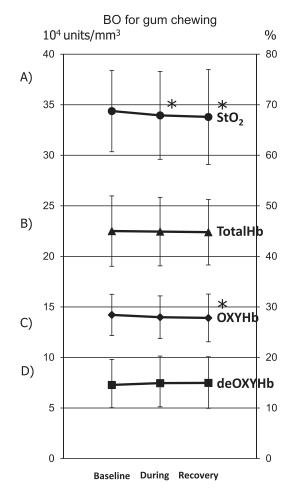
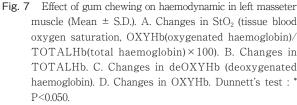


Fig. 6 Electromyographic (EMG) activity and changes of blood oxygenation (BO) during gum chewing from one subject (Subject No. 11).





# DISCUSSION

The main findings of this study was that an experimental tooth grinding task did not cause significant changes in hemodynamic characteristics of the masseter muscle in contrast to a static maximal contraction task and a rhythmic dynamic contraction task.

In this study, maximal voluntary contraction was used as one of the standardized oral-motor tasks in order to see the changes of intramuscular BO in masseter. The results showed that the isometric contraction task evoked the slight increase of both oxygenated and deoxygenated haemoglobin and resulted in the significant increase of total haemoglobin. It is suggestive that isotonic masseter muscle contractions are associated with increased intramuscular blood flow in the masseter muscle compared to that during rest (Baseline). This point is in good agreement with thea previous study<sup>12</sup>). Furthermore, the current masseter BO data did not show theany reactive hyperaemia, which can normally be seen in limb muscles after MVC tasks<sup>16, 21</sup>. The reactive hyperaemia has been thought to be a critical factor for assessing and recognizing if the blood flow is sufficient to meet metabolic demand<sup>10</sup>. It seems that the characteristics of BO in masticatory muscle isare different from thatthose in the limb muscles.

The current tooth-grinding task did not cause the changes in any of masseter muscle blood oxygenations. However, in fact, raw data (Fig. 4) showed the repetition of increases and decreases (waves) of oxygenateddeoxygenated-, and total-haemoglobin levels and the decreases of StO2 levels taking accompanied with the increase and decrease of electromyographic activities during the task. This means that eccentric tooth grinding, actually, causes the substantial changes on masseter muscle blood oxygenations. For the future study, the new technique for analyses, which can explain these small changes, would be needed. However, the present results showed that bruxism-like tooth-grinding activity does not seem to lead to the muscle pain condition such as an energy crisis with release of metabolites and neuroactive substances.

Finally, there is a new finding that the current gumchewing task was associated with a significant decrease of StO2 during the task and lasted into the post task period, which may due to the decreased levels of OXYHb. The current results appear to be different from a previous study<sup>22)</sup>, which adopted a 6-min gumchewing task and did not find changes in OXYHb. This finding may therefore support the theory that masseter haemodynamics during chewing movements involves muscle pumps accompanied by the rhythmic contraction of the muscles. The possible reason for this discrepancy is that, in the present study, the BO recording was performed when the texture of chewing gum had become soft after about 2 min. Thus, the current results are only reflecting the BO changes between 2 to 3 min of chewing whereas the previous study examined the entire 6 min task. It may be interesting to follow the time course more in detail when future studies on masseter muscle BO are planned.

Bruxism has for a long time been thought to be an important aetiologic factor for temporomandibular disorder including POMP<sup>1-3)</sup>. Overall, the present

results do not point towards major disruption of the haemodynamic function during the tooth-grinding task but it may be premature to exclude the possibility that longer-duration tooth grinding or perhaps higher-intensity tooth grinding may trigger vascular responses that could contribute the pathophysiology of TMD pain and POMP. Further studies will be needed to address this important issue.

# CONCLUSION

These results showed that high-intensity experimental tooth clenching caused constriction-like reactions in the masseter muscle whereas tooth grinding did not cause detectable changes in haemodynamic characteristics of masseter muscle. Finally, the findings indicated that rhythmic dynamic contractions might lead to oxygen deficit in the masseter muscle. The present data may have implications for understanding the potential pathophysiological consequences of different types of oralmotor tasks, e.g., bruxism and prolonged mastication.

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