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Human Movement Science xxx (2015) xxx-xxx

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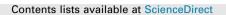
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Acute effects of jaw clenching using a customized mouthguard on anaerobic ability and ventilatory flows

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ABSTRACT

The latest findings on the ergogenic effects of a dentistry-design, bite-aligning mouthpiece require additional research to assess its impact on anaerobic ability and ventilatory parameters. This paper was aimed at determining the ergogenic acute effects of wearing a custom-made mouthpiece on oral airflow dynamics, 30-s Wingate Anaerobic Test performance parameters.

Twenty-eight healthy and physically-active male subjects (age: 24.50 ± 3.32 , height: 181.34 ± 7.4 , weight: 78.14 ± 8.21), were voluntarily studied. The subjects were first briefed on the test protocols, and then performed the 30 s Wingate test and Spirometer test. The experimental trials were performed in a random counterbalanced order. We evaluate maximum expiratory volume ($V_{\text{Emax}} \text{Lmin}^{-1}$), mean power (W kg⁻¹), peak power (W kg⁻¹), time to peak (s), rate to fatigue (W s⁻¹) and lactate production (mMol L⁻¹), rate of perceived exertion (RPE).

There were significant differences between mouthguard and no-mouthguard conditions in mean power (W kg⁻¹), peak power (W kg⁻¹), time to peak (s), and rate to fatigue (W s⁻¹) for the 30-s Wingate Anaerobic Test. Significantly lower lactate production (mMol L⁻¹) was observed, in mouthguard condition but no significant differences were found in RPE. In airflow dynamics, the $V_{\rm Emax} L \min^{-1}$ was significantly higher when comparing the mouthguard and the no mouthguard conditions in both forced and unforced conditions.

In conclusion, wearing a customized mouthguard improves anaerobic ability and increases forced expiratory volume. This study will help practitioners improve athlete's performance in anaerobic activities where high intensity action might provoke jawclenching, contributing in reductions of lactate and fatigue, and improving ventilatory parameters.

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51 1. Introduction 52

53 In some sports, athletes use different types of mouthguards to prevent dental injuries and to protect the maxillofacial 54 structure from possible shocks and collisions. Mouthguards allow athletes to feel protected, but the devices are not always 55 comfortable. There are three main types of mouthguards: standard, self-adapted, and customized. Standard mouth guards 56 are a fitted piece that requires no adjustments. Due to its relatively low cost, it is the most commonly used mouthguard,

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22 September 2015

J. Morales et al./Human Movement Science xxx (2015) xxx-xxx

but is often uncomfortable. Self-adapted mouthguards consist of a "thermoplastic" liner that can be manipulated with heat during an at-home fitting process to improve the fit to the maxillary teeth. This type is also inexpensive and widely available. Finally, customized mouthguards require dental impressions or use of a scanning process of the dental structure of an individual's teeth. This is the most expensive of the three types of mouthguards and often requires the expertise of a dentist (Chalmers, 1998; Geary, Clifford, & Kinirons, 2009; Patrick, van Noort, & Found, 2005). Beyond the preventive role of wearing mouthguards, recent research has investigated its physiological effects and the possible benefits in different parameters.

63 Although it has been demonstrated that wearing a mouthguard reduces orofacial injury, many athletes do not wear them 64 during training sessions or in competition for different reasons, including speech and breathing difficulties or discomfort 65 (Collares, Correa, Da Silva, Hallal, & Demarco, 2013; Duarte-Pereira et al., 2008; Gebauer, Williamson, Wallman, & Dawson, 2011). In recent years, several researchers have studied the effects of wearing mouthguards on physiological and 66 breathing parameters in an attempt to demonstrate that they are unlikely to interfere with an athlete's performance. 67 68 Francis and Brasher (1991) studied the effect of three different types of mouthguards on forced expiratory volume, maximum flow rate in a single forced expiration, subject's VO₂max, and one-minute ventilation of expired gases collected during 69 70 a cycling exercise. The authors found that wearing mouthguards produced an effective respiratory pattern during brief periods of heavy exercise, which might improve tissue oxygenation and lower metabolic cost (Francis & Brasher, 1991). Amis 71 et al. (2000) studied the influence of intra-oral maxillary sports mouthguards on the airflow dynamics of oral breathing 72 and found that mouthguards cause obstruction to oral breathing, mainly at rest and when the degree of mouth opening 73 74 is restricted; however, these are unlikely to cause interference at high ventilatory rates (Amis, Di Somma, Bacha, & Wheatley, 2000). Other studies have found no difference in oxygen consumption and ventilation in trained subjects 75 (Bourdin et al., 2006; Collares et al., 2013; Gebauer et al., 2011; Rapisura, Coburn, Brown, & Kersey, 2010). 76

77 For some sports, ventilatory mechanisms are closely connected to the anaerobic recovery phases. Thus, wearing a mouth-78 guard wouldn't hamper oxygen intake during these phases. Furthermore, mechanisms like concurrent activation potentia-79 tion (CAP) that are derived from a clenched jaw and which constitute a remote voluntary contraction (RVC) have been 80 suggested as the possible causes of an ergogenic effect of wearing mouthguards (Ebben, 2006; Ebben, Flanagan, & Jensen, 81 2008). The neuromuscular effects of jaw-repositioning and contraction of the mandible muscles may translate to improved neuromuscular responses in active exercise movers. As a consequence, the activation of different limb muscles contribute to 82 strengthen movements like rowing, pedaling, running, or jumping (Milani, De Periere, Lapeyre, & Pourreyron, 2000). When 83 one part of the motor cortex is active, connections to other areas of the motor cortex are also affected (Ebben, 2006). Thus, 84 85 CAP has been suggested as a performance enhancer with regards to several acute power and strength tasks in sports thanks to the RVC of jaw muscles (Ebben, 2006; Ebben, Leigh, & Geiser, 2008; Forgione, Mehta, & Westcott, 1991). For instance, ana-86 lyzing the muscle activation via electromyography in a group of healthy and active men and women, the muscles involved in 87 a RVC are more active; this increase in activity results in a greater activity in the prime movers in isokinetic knee extension-88 flexion (Ebben, Kaufmann, Fauth, & Petushek, 2010). Furthermore, it has been suggested that biting a custom made mouth-89 90 guard could enhance athletic performance in quick powerful and anaerobic actions and increase ventilatory pathways. Garner and McDivitt (2009) used computer tomography scans and found that the use of a jaw-repositioning mouthpiece 91 92 increased oropharynx width and diameter, and concluded that the increased airway opening promoted improved gas exchange in endurance athletes. Arent et al. (2010) compared the effects of a non-dentistry design mouthguard and a tra-93 94 ditional custom-fitted mouthguard and found significant differences in countermovement vertical jump (CMVJ) height and peak power in a 30 s Wingate Anaerobic Test (WANT). They used transcutaneous electrical nerve stimulation (TENS) 95 96 to ensure relaxation of the lower jaw before the fitting process of the mouthguards. Cetin et al. (2009) also found significant 97 increases in relative peak and average power in WAnT for elite taekwondo athletes who wore a mouthguard, compared to those who wore no mouthguard. Dunn-Lewis et al. (2012) compared the effects of three conditions (customized, self-fitted, 98 99 and no mouthguard) on different power performance tests. They reported significantly higher power production in plyo press power quotient (i.e., a more specific anaerobic performance test than WANT) and higher rates of power development 100 in CMJV for men who used a customized mouthguard. In contrast, Bourdin et al. (2006) found no significant differences in 101 force and power parameters in a cycle ergometer, for trained men who used customized and self-adapted mouthguards. 102 Other studies found no significant improvements of performance in anaerobic ability tests (Bailey et al., 2014; Duarte-103 104 Pereira et al., 2008; Duddy et al., 2012).

The latest findings on the ergogenic effects of a dentistry-design, bite-aligning mouthpiece require additional research to assess its impact on anaerobic ability and ventilatory parameters. Therefore, this study sought to investigate the acute effects of a customized, bite-aligning mouthpiece on the different parameters assessed during a WnAT, as compared to clenching the jaw without a mouthpiece, in a physically-active, male population. The study also investigated the influence of wearing a mouthpiece on non-forced and forced airflow dynamics, as compared to open mouth and a jaw clenching without mouthpiece. Mouthpieces were made using a new scanning method that simplified the fitting process and lowered its cost.

111 2. Methods

Twenty-eight healthy and physically-active male subjects (age: 24.50 ± 3.32, height: 181.34 ± 7.4, weight: 78.14 ± 8.21, body mass index: 24.81 ± 1.47), were voluntarily recruited for this study. All participants were physically active in different disciplines (from 3 to 4 training sessions per week of soccer, handball, basketball or field hockey), competed in regional

22 September 2015

J. Morales et al./Human Movement Science xxx (2015) xxx-xxx

championships and were free of temporomandibular joint disorders. The subjects were required to stop training sessions at least 24 h before the testing sessions. They were also asked not to intake any stimulating substance (caffeine, theine...) and alcohol drinks one day before. The study was performed with informed consent and following all the guidelines for experimental investigation with humans. The ethics committee of the Ramon Llull University of Barcelona approved the development of this study which was conducted according to the latest version of declaration of Helsinki.

120 2.1. Experimental overview

Each subject participated in three sessions. The first session was used to assess anthropometric measurements and to scan the subject's mouth structure. In this session, an expert dentist evaluated the possible temporomandibular joint disorders and the quality of occlusion. Subjects with a consistent malocclusion were excluded from the study. In the second session, subjects were familiarized with the test protocols via a learning session that included demonstration of WnAT and the airflow dynamics measurements. Subjects also performed the airflow tests and the first WAnT trial. In the third session, subjects performed the second WnAT trial. Conditions were randomly distributed in all tests. WnAT trials were separated by three days and subjects were not allowed to perform intensive training workloads.

The intraoral device CleverBite® (Cleverbite SL, Terrassa, Spain) is a mouthguard Class III (American Society for Testing & 128 Materials, 2000), and is manufactured by taking a digital recording of a scan of both the maxillary and mandibular dental 129 130 arches (Fig. 1). This record is complemented by a digital recording of the interocclusal relation associated with the resting position of the mandible. The device fabrication is performed on a digital model made of Pearlstone VisiJet Plastic Material 131 (Urethane Acrylate and Phenylbis (2, 4, 6-trimethylbenzoyl) phosphine oxide) obtained by scanning the dentoalveolar max-132 133 illary and mandibular environment using the 3Shape Trios System (3Shape Inc. Copenhagen, Denmark). Prior the scanning, 134 subjects performed different maneuvers to neutralize any postural neuromuscular disorders that might influence the centric 135 mandible position with respect to cranio and the cervical muscle activation. Subjects wore a 2-cm long and 0.5-cm thick 136 cylindrical piece of cotton in both sides of the mouth (i.e., at the level from first molar to the distal slope of the canine 137 cusped) and were asked to walk barefoot for 2 min. After walking, subjects performed neck rotations, flexo-extensions, and lateral flexions for 1 min each. 138

139 2.1.1. Wingate Anaerobic Test

140 Anaerobic power and capacity were assessed using the 30-s WAnT with and without mouthguard (MOUTHG and NO-MOUTHG, respectively). Subjects completed a 30-s maximal effort on an electronically braked cycle ergometer Lode Excal-141 ibur Sport V2.0 (Lode Medical Technology, Groningen, The Netherlands) at a resistance equivalent to 7.5% of their body mass. 142 The ergometer was interfaced with a computer loaded with software (Wingate Software Version 1.11, Lode BV) that applied 143 the appropriate load for each subject. As a warm-up procedure, subjects were instructed to begin pedaling for 5 min at 144 100 W and approximately 60 rpm. After a 5 s countdown and without altering the mentioned parameters, subjects were 145 asked to begin pedaling as fast as possible while receiving verbal encouragement throughout the test. Peak power, mean 146 power, minimum power, time to peak power, relative peak power, relative mean power (W) and fatigue index (W s⁻¹) were 147 calculated and recorded in an online data acquisition system. The rate of perceived exertion (RPE) was collected after each 148 149 trial using a modified 10-point Rating of Perceived Exertion Scale (CR-10:RPE) (Borg, Hassmén, & Lagerström, 1987). Capil-150 lary blood samples were taken on right fingertips at the third minute after completion of the test and analyzed for lactate concentration by using the Lactate Pro (LT-1710, Arkray, Japan) portable analyzer. 151



Fig. 1. Type of mouthguard used for the study.

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22 September 2015

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4

J. Morales et al./Human Movement Science xxx (2015) xxx-xxx

152 2.1.2. Airflow dynamics measurements

While in standing position, subjects were asked to breath at two different paces (NON-FORCED and FORCED) during 30-s 153 under three different conditions: open mouth without mouthguard, jaw clenching without mouthguard and jaw clenched 154 155 with mouthguard (OMNM, ICNM and ICM, respectively). In NON-FORCED breathings, the subjects were asked to breath in 156 a resting pace (approximately at 30 inspirations per minute). In FORCED condition, subjects were asked to breath as quick and deep as possible. Rest time was 3 min after both measurements under each pace and 6 min between conditions. Con-157 158 ditions were randomly distributed. A AFT21 non-rebreathing T valve and a AFT9 mouthpiece with the AFT4 bacterial filter 159 were mounted on a AFT25 facemask and was connected to a Biopac MP100 system through the wide-range airflow trans-160 ducer amplifier TSD107B (Biopac Systems, Inc., CA, United States). The system recorded variations in airflow during the exercise, and the software Acknowledge 3.0.9. (Biopac Systems, Inc., CA, United States) plotted and recorded the flows on a time 161 scale. The sample rate was established at 200 Hz. Peak air flow was detected and mean air flow during the 30 s interval was 162 calculated for all conditions. 163

164 2.2. Statistical analysis

Statistical analyses were performed with the Statistical Package for Social Science version 22.0 (SPSS, Inc., Chicago, IL). The Shapiro–Wilks test was used to test for normal distribution of the data. To compare Wingate parameters according two conditions, a paired *t*-test was used. When the data did not follow a normal distribution, an equivalent nonparametric test was applied. Comparisons between the three different conditions for airflow dynamics were carried out using a mixed model for repeated measures ANOVA. A significance level of p < .05 was used for all tests.

170 3. Results

The mean contrast for dependent WAnT variables was tested through t-test for related samples. Results showed a significantly higher performance (P < 0.05) of MOUTHG in all variables (Table 1).

The RPE data was not normally distributed and a non-parametric Wilcox test was used to test the mean differences. Results showed no significant differences (P < 0.05, W = 0.84) between NO-MOUTHG (7.89 ± 0.83) and MOUTHG (7.75 ± 0.75).

176 Mean contrasts for airflow dynamic variables was tested using a mixed model of two-way repeated measures ANOVA. A 177 within-subject analysis compared the performance of the three conditions (OMNM, JCNM and JCM) \times 1 type of breathing 178 between-factor (FORCED and NON-FORCED). When a significant *F* score was found, Bonferroni *post hoc* tests were used to 179 test the pairwise differences between the performances under the three conditions.

Multivariate analysis of mean airflow showed a significant main effect for condition ($F_{(2,53)} = 47.85$, p < 0.05; $\eta_p^2 = 0.64$) and type of breathing ($F_{(2,53)} = 38.75$, p < 0.05; $\eta_p^2 = 0.59$). Univariate contrast show a significant main effect for condition on type of breathing ($F_{(1.89,97,21)} = 49.77$, p < 0.05; $\eta_p^2 = 0.48$). In this case, the sphericity assumption was violated and the number of degrees of freedom was adjusted using the Huynh–Feldt method. Also, the analysis of peak airflow show a significant main effect for condition ($F_{(2,53)} = 54.64$, p < 0.05; $\eta_p^2 = 0.67$) and type of breathing ($F_{(2,108)} = 39.80$, p < 0.05; $\eta_p^2 = 0.60$). Univariate contrast show a significant main effect for condition on type of breathing ($F_{(2,108)} = 34.67$, p < 0.05; $\eta_p^2 = 0.39$).

Pairwise comparisons indicated significantly better performance in mean airflow for FORCED with respect to NON-FORCED for the three conditions (P < 0.05). No significant differences were found between the three conditions in NON-FORCED (P > 0.05). However, in FORCED, a significantly higher air volume was expired in subjects using OMNM as compared to JCNM and JCM, and significantly higher air volume was expired in JCNM condition (P < 0.05) (Fig. 2).

There were significant differences in FORCED peak airflow for OMNM as compared to JCM and JCNM (P < 0.05). In contrast to mean airflow, there were no significant differences between JCM and JCNM (P > 0.05).

Power analyses were performed for all the variables after participant recruitment using GPOWER software (Bonn FRG,
 Bonn University, Department of Psychology). The *post hoc* analysis (given α, sample size, and effect size) demonstrated a
 power of 99%.

Table 1

Comparisions of Wingate variables wearing mouthguard and no mouthguard.

Wingate variables	No Mouthguard Mean (SEM)	With Mouthguard Mean (SEM)	t ₍₂₇₎	r
Mean power BW (W kg ⁻¹)	9.01 (.15)	9.14 (.15)	-2.55	.44
Peak power BW (W kg ⁻¹)	16.04 (.47)	16.69 (.43)	-2.91	.48
Time to peak (s)	4.55 (.08)	4.2 (.05)	3.07	.51
Fatigue (W s ⁻¹)	31.10 (1.53)	28.32 (1.41)	2.93	.49
Lactate (mmol L ⁻¹)	11.91 (.34)	11.01 (.35)	2.35	.45

Abbreviations: BW = body weight; SEM = standard error of measurement; $t_{()} = t$ -test value(degree of freedom); r = statistical power.

* Indicates a significant differences between the two conditions.



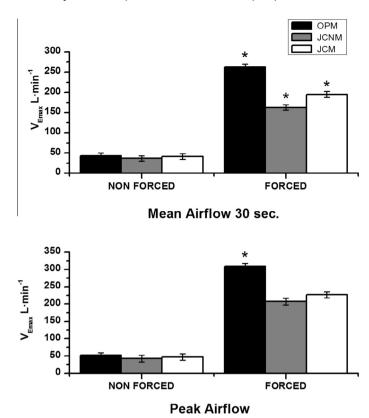


Fig. 2. Comparisons airflow measuraments between the three conditions. ONM = Open mouth without mouthguard, JCNM = Jaw clenching without mouthguard, ¹Indicates a significant differences between the three conditions (P < 0.05).

195 4. Discussion

This study found significant differences in all variables of performance in WAnT. Subjects reached higher performances in MOUTHG condition for mean power body weight (BW $[W kg^{-1}]$), peak power BW $(W kg^{-1})$, time to peak (s) and fatigue (W s⁻¹). Also, subjects experienced significantly lower lactate accumulation (mMol L⁻¹) under MOUTHG as compared to NO-MOUTHG conditions, but similar RPE. These findings may hold practical relevance for athletes whose sports require anaerobic effort, and especially for those athletes who need mouth protection.

The anaerobic ability test results of this study align with Arent, McKenna, and Golem (2010) which reported significant 201 differences in WAnT peak and average power for athletes wearing a neuromuscular, dentistry-design mouthguard respect 202 203 standard custom-fitted mouthguard condition. Arent et al. did not perform a comparison between an open-mouth condition 204 and jaw clenching condition but indicated acute ergogenic effects caused by the neuromuscular mouthguard. This study did not use TENS to ensure relaxation of the lower jaw before the mouthguard fitting process; however some relaxation tech-205 niques were used. These previous mechanisms to improve the mouthguard fitting process improved the adjustability of the 206 207 mouthpiece and may have served as a possible performance enhancer factor. Peak power and average power of the WAnT 208 were also reported in similar studies (Cetin, Kececi, Erdòan, & Baydar, 2009; Jung, Chae, & Lee, 2013). The effects of a RVC on 209 the promotion of CAP is a possible explanation of the enhanced performance on anaerobic ability (i.e., both lactic and alactic). 210 Peak and time to peak are consistent indicators of alactic anaerobic power and correlated with other power indicators such as CMVJ. Some studies reported ergogenic benefits of wearing mouthguards on jump performance (Ebben et al., 2008, 2010). 211 212 Average power and fatigue index are used to explain the lactic anaerobic power of the subjects. Other studies have also reported ergogenic effects of wearing a mouthguard as compared to no mouthguard on different tests of lactic anaerobic per-213 214 formance in different populations (Cetin et al., 2009; Duddy et al., 2012; Dunn-Lewis et al., 2012). Milani et al. (2000) support 215 the idea that jaw repositioning improves functional proprioception and promote benefits in movement and performance. 216 According to Ebben et al. (2008) the combination of jaw clenching and RVC of lower body muscle groups increase the rate of force development and decrease blood lactate production. These conditions could enhance pedaling efficiency increasing 217 218 the reflex activity of the Soleus, without energy expenditure, and reducing lactate production (Ebben et al., 2008). Further-219 more, the improvement on airflow during jaw clenching can increase aerobic metabolism in reduction of anaerobic lactate 220 production (Garner & McDivitt, 2009).

22 September 2015

6

J. Morales et al./Human Movement Science xxx (2015) xxx-xxx

While other studies have also reported differences between traditional boil-and-bite and custom-fitted mouthguards (Dunn-Lewis et al., 2012). In the present study, one limitation is that only the differences between mouthguard and nomouthguard conditions were established. No comparison between different types of mouthguard was done. However, in this study should be noted that the mouthguard fitting process provide a precise adjustment and comfortable design and resulted in good conditions for powerful jaw clenching and taking benefits of the CAP. Indeed, the RVC of the jaw muscles seems to be facilitated by wearing a custom-fitted mouthguard and this consist a mechanism for CAP promotion. Therefore, the activation of muscles involved in powerful movements may be enhanced by CAP.

Exposing subjects to a 30 s maximal effort on a braked cycle ergometer produces a considerable amount of lactate that then accumulates in blood. Although subjects demonstrated significantly higher average power and lower fatigue, significantly lower blood lactate accumulations were found for subjects wearing a mouthguard. This finding is consistent with Bailey et al. (2014) and indicates that mouthguard use might create an advantage for sports where fatigue and lactate accumulations affects athletic performance. In accordance with the aforementioned study, we also did not found statistically differences in RPE.

234 This study found significant differences in airflow dynamics measurements (i.e., 30 s forced breathings under the same condition). To elucidate the potential mechanism involved with mouthguard use during intense exercise, patterns of 30 s 235 mean airflow and peak airflow were assessed. This study's results differ from those of other studies that found no significant 236 differences between use and no use of the two types of mouthguard on ventilatory parameters. This study found that both 237 238 mean and peak airflow had a significant increase in JCM condition, as compared to JCNM and OMNM. Although prior studies 239 used incremental tests, this study found similar mean airflow values at maximal intensities when comparing the values of FORCED condition with JCM and JCNM (Gebauer et al., 2011; Rapisura et al., 2010). These studies also observed no significant 240 differences in ventilatory parameters between wearing and not wearing mouthguards under low intensity conditions; this 241 242 study showed similar results.

243 This study showed how the wearability and comfort of the mouthguards improved forced expiratory airflow; these 244 mouthguards could help to prolong exercise duration by improving ventilation and effort efficiency, as has been found in 245 other studies (Garner, Dudgeon, & Mcdivitt, 2011; Garner, Scheett, & Mcdivitt, 2011; Rexhepi & Brestovci, 2013). Moreover, Duarte-Pereira et al. (2008) showed that custom-fitted mouthguards have less interference with airflow than a boil-and-bite 246 247 self-adapted mouthguards. According to the findings in this study, this contributes to the idea that custom-fitted mouthguards interfere less in ventilation. The respiratory patterns observed in this study showed that there are significant differ-248 249 ences between jaw-clenching forced breathing wearing mouthguard respect non wearing it, assuming that open mouth situation offered better airflow dynamics (Garner, Scheett, & McDivitt, 2011). A possible explanation for the improved ven-250 tilation during jaw clenching with mouthguard may be that custom mouthpieces improve jaw position by increasing 251 oropharynx width and diameter (Garner, Dudgeon, & McDivitt, 2011). 252

Further investigation is needed to compare use of different type of mouthpieces. Moreover, other athletic performance parameters should be tested in different sports and in women. Also, data of jaw clenching force might clarify the influence of different RVC magnitudes on the forces exerted by different prime movers in sports (Ebben et al., 2010). These findings could help practitioners improve an athlete's performance, particularly in sports where there are high-intensity anaerobic efforts that where high intensity action provokes jaw-clenching, thereby reducing lactate production and fatigue.

In conclusion, this study demonstrated that the use of custom-made, bite-aligning mouthguard had an ergogenic effect on WANT variables, lactate production, and airflow dynamics in healthy young men compared with those who did not wear a mouthguard. This has important implications for athletes engaged in anaerobic efforts that cause jaw-clenching, and for those who need to reduce the incidence of dentoalveolar trauma.

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22 September 2015

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