

# Short-Term Sensorimotor Effects of Experimental Occlusal Interferences on the Wake-Time Masseter Muscle Activity of Females with Masticatory Muscle Pain

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**Aims:** To investigate the effects of the application of an acute alteration of the occlusion (ie, interference) on the habitual masseter electromyographic (EMG) activity of females with temporomandibular disorders (TMD)-related muscular pain during wakefulness. **Methods:** Seven female volunteers with masticatory myofascial pain participated in a crossover randomized clinical trial. Gold foils were glued on an occlusal contact area (active occlusal interference, AI) or on the vestibular surface of the same molar (dummy interference, DI) and left for 8 days. The masseter electromyogram was recorded during wakefulness in the natural environment by portable recorders under interference-free, dummy-interference, and active-interference conditions. The number, amplitude, and duration of EMG signal fractions with amplitudes above 10% of the maximum voluntary contraction (activity periods, APs) were computed in all experimental conditions. Muscle pain, headache, and perceived stress were each assessed with a visual analog scale (VAS), and an algometer was used to assess masseter and temporalis pressure pain thresholds. Data were analyzed by means of analysis of variance. **Results:** The frequency and duration of the recorded APs did not differ significantly between the experimental conditions ( $P > .05$ ), but a small and significant reduction of the EMG mean amplitude of the APs occurred with AI ( $P < .05$ ). Neither the VAS scores for muscular pain, headache, and perceived stress nor the pressure pain thresholds changed significantly throughout the entire experiment ( $P > .05$ ). **Conclusion:** An active occlusal interference in female volunteers with masticatory muscle pain had little influence on the masseter EMG activity pattern during wakefulness and did not affect the pressure tenderness of the masseter and temporalis. *J Oral Facial Pain Headache 2015;29:331–339. doi: 10.11607/ofph.1478*

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Occlusal interferences have been considered as a risk factor for the development of temporomandibular disorders (TMD)<sup>1,2</sup> and have been widely investigated in human and animal studies.<sup>3–13</sup> Human studies (for review, see Clark et al<sup>14</sup>) have indicated that the application of experimental occlusal interferences may enhance the risk of developing TMD by increasing masticatory muscle activity, which in turn may lead to muscle overload and pain.<sup>6,15,16</sup> This hypothesis also has been confirmed in animal models, where occlusal interferences have been shown to produce brainstem immunohistochemical changes, trigeminal central sensitization, long-term masticatory muscle hyperalgesia, and facial hypersensitivity.<sup>17–18</sup> In contrast with these findings are those of a clinical trial reporting that healthy female volunteers do not develop signs and/or symptoms of TMD after the placement of an experimental occlusal interference.<sup>11</sup> Most participants adapted fairly well to the occlusal disturbance, showing a decrease of masseter muscle contractions<sup>11</sup> and no changes of pressure pain thresholds of the masticatory muscles.<sup>19</sup>

It has been reported that the response to occlusal interferences may be different between individuals with or without TMD.<sup>20,21</sup> Indeed, previous studies carried out in individuals without TMD found that the introduction of experimental occlusal interferences may cause transient

tooth pain as well as signs and symptoms of TMD, which disappear in 1 to 2 weeks.<sup>3-9</sup> Conversely, individuals with a former TMD history seemed to adapt less well to an experimentally placed occlusal interference than TMD-free controls, as assessed by patient reports.<sup>22</sup> However, previous research has not assessed the electromyographic (EMG) activity of masticatory muscles under the experimental occlusal interference condition. Hence, the aim of this study was to investigate the effects of the application of an acute alteration of the occlusion (ie, interference) on the habitual masseter EMG activity of females with TMD-related muscular pain during wakefulness. The effects of the interference on TMD-related pain symptoms as well as on pressure pain thresholds of the masticatory muscles were also investigated. It was hypothesized that the insertion of an experimental occlusal interference would change the contraction pattern of the masseter muscle during wakefulness and pressure sensitivity of the jaw elevator muscles in a group of females affected by myofascial pain.

## Materials and Methods

### Subjects

Only women were recruited for this research. A flyer was disseminated to 86 female medical students at the University of Naples Federico II. It included the question, "Have you had pain in the face, jaw, temple, in front of the ear or in the ear in the past month?" (Question #3, Axis I, Research Diagnostic Criteria for TMD [RDC/TMD]<sup>23</sup>). Students who answered positively were invited to participate in this research project.

Eighteen out of the 86 female students reported orofacial pain and expressed an interest for participation. The students were extensively informed about the experimental procedures, the duration of the trial, and possible unwanted effects, and they were told that they could leave the study at any time.

These subjects underwent a preliminary clinical examination at the Section of Temporomandibular Disorders and Orofacial Pain of the University of Naples Federico II, Italy, with an examiner (AM) calibrated according to the RDC/TMD.<sup>23</sup> The RDC/TMD criteria defined in 1992 were used because the study was performed before the publication of the new diagnostic criteria for TMD.<sup>24</sup> Psychosocial evaluation was performed according to Axis II-RDC/TMD (Graded Chronic Pain Scale [GCPS] and Symptom Checklist-90-Revised [SCL-90-R]).<sup>23</sup> The participants were also invited to score their perceived facial pain on a numeric rating scale by answering the question, "How would you rate your facial pain on a 0 to 10 scale at the present time, that is, right now,

where 0 is "no pain" and 10 is "pain as bad as could be"? (Question #7, Axis I, RDC/TMD<sup>23</sup>). Only individuals with a diagnosis of masticatory muscle pain, ie, myofascial pain (RDC/TMD Ia/Ib) were selected for the study. Exclusion criteria were previous TMD examination and/or treatment, inflammatory conditions, periodontal disease, dental prostheses, occlusal wear (> 2 as defined by Clark et al<sup>25</sup>), previous orthodontic treatment, absence of one or more teeth with the exception of third molars, single-contact balancing side and protrusive occlusal interferences, slide from retruded contact position to intercuspal position greater than 2 mm, neurological disorders, nail biting, smoking, and habitual drug intake.

Ten participants were excluded according to the inclusion/exclusion criteria. The final sample included eight female volunteers suffering from myofascial pain. None of them reported a history of sleep bruxism, and no participant was using birth control pills at the time of the experiment. All participants signed an informed consent form and were compensated with 150 euros for participation in the study. The study protocol had been approved by the local Ethics Committee (protocol number #13900–University of Naples Federico II, Italy).

### Study Design

The study set-up was similar to that of a previous study performed in TMD-free women.<sup>11</sup> It was carried out in a double-blind crossover design, with participants serving as their own control. Each subject went through four different study conditions during a 6-week period: interference-free condition, eg, before interference application (IFC<sub>before</sub>); dummy interference condition (DIC); active interference condition (AIC); and interference-free condition after interference removal (IFC<sub>after</sub>). Active and dummy interferences were left in place for 8 days (Fig 1).

The order of interference application, either the AIC or DIC first, was determined by means of a balanced block randomization.

### Occlusal Interference

Dental impressions were taken of both dental arches (Palgat Plus, 3M Unitek) and poured with stone (Elite Master, Zhermark). The casts were mounted in the intercuspal position in a semiadjustable articulator (Panadent 1210; Panadent Co). The subject's contact points between the maxillary and mandibular first molars when she was closing in the intercuspal position were marked using marking paper (Accufilm II, Parkell) and then reproduced on the stone casts with a pencil.

A gold foil strip (length × width × height = 2.0 × 8.0 × 0.2 mm; weight = 0.05 g) was glued to the mandibular first molar of the preferred chewing side (right side for six subjects and left side for two subjects)

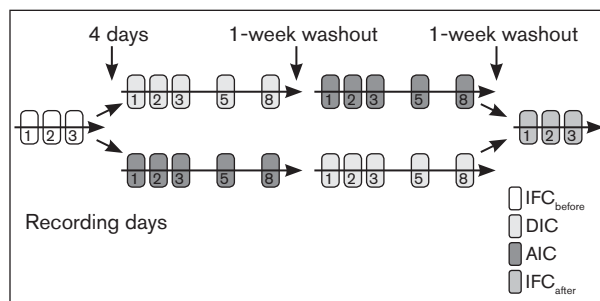
in the area of the occlusal contact and carefully adapted to the tooth anatomy on dental casts (active interference). The gold foil strip disturbed the intercuspal position but did not interfere with occlusion during lateral or protrusive mandibular movements. To create the dummy interference, a second strip was placed on the vestibular surface of the same tooth without interfering with the intercuspal position. The active and the dummy interferences were glued to the tooth by means of composite (Revolution, Kerr) by one of the authors (RM) at AIC<sub>day-1</sub> and DIC<sub>day-1</sub> respectively.

### Surface Electromyography and Data Analysis

The EMG activity of the masseter muscle ipsilateral to the interference side was recorded by means of portable EMG recorders.<sup>26,27</sup> These also measured the impedance between the electrodes every 5 minutes in order to alert the participants with a beep in case of disconnection. Two surface EMG electrodes (model 13L20, Dantec; 6 mm diameter) were attached to the skin overlying the masseter muscle always by the same examiner (PF), who was unaware of the occlusal interference condition. The electrodes were placed on the preferred chewing side and aligned along the muscle fiber direction with a distance of 20 mm center to center as in a previous study.<sup>27</sup> The reference electrode was attached to the skin overlying the mastoid process ipsilateral to the recording side. Prior to attaching the electrodes, the skin was rubbed with alcohol-soaked gauze. Accurate relocation of the electrode sites was achieved using transparent plastic templates.<sup>28</sup> The first 3 minutes of each EMG recording included the following standardized tasks: 3 maximum voluntary contraction (MVC) tasks with a 10-second interval; 3 saliva swallowing; and 20 simulated artifacts, eg, pulling and touching the electrodes, each 10 times in order to avoid having the noise created by artifacts recorded as EMG activity.

The individual EMG amplitude during MVC was calculated by averaging the mean maximum EMG values of the six recordings performed before the interference-free conditions (IFC<sub>before</sub>, IFC<sub>after</sub>) (Fig 1). The EMG signal analysis included calculation of the number of contraction episodes, ie, activity periods per hour (AP/h), their net duration (Dur), and mean amplitude (A<sub>mean</sub>). An activity period corresponded to a portion of the signal above a predetermined threshold, which could contain subthreshold signal portions shorter than a predetermined standby time of 5 seconds.<sup>27</sup> The threshold to determine an AP was set at 10% MVC, as in a previous study.<sup>11</sup>

The recording time was set between 10 am and 7 pm. Subjects were asked to eat between 1 pm and 2 pm; to avoid gum chewing, physical exercise, sleeping, and electromagnetic fields; and to return the recorder the following day for offline analysis.



**Fig 1** Experimental design. IFC<sub>before</sub> = interference-free condition before gold foil application; AIC = active interference condition; DIC = dummy interference condition; IFC<sub>after</sub> = interference-free condition after gold foil application.

### Clinical Protocol

After 3 consecutive days of baseline EMG recordings (IFC<sub>before</sub>), the active or dummy interference was applied, with each subject being assigned to two treatment sequences including both AIC and DIC. Each interference was left in place for 8 days. This time window was the same used in previous studies and was long enough to include nonworking days as well. The treatment sequences were prepared and concealed before assignment of the individual sequence. The masseter EMG activity was recorded during 5 days, ie, at days 1, 2, 3, 5, and 8. After a 1-week washout period, another foil strip was attached; participants who received an active interference first received a dummy interference and vice-versa. After the second washout period, the EMG activity was monitored for 3 additional consecutive days, again with the teeth free of any foil strips (IFC<sub>after</sub>). A total of 16 recording days was obtained from each subject (Fig 1).

### Subjective Assessments

Facial pain, headache, current stress, and occlusal discomfort were each assessed by means of a 100-mm visual analog scale (VAS).<sup>29</sup> The left endpoint of the scales indicated “no pain/headache/stress/discomfort at all” and the right endpoint corresponded to “worst imaginable pain/headache/stress/discomfort.” VAS ratings were collected at the beginning of each recording day.

The clinical examination was performed by the same blind examiner (AM) at the following days: IFC<sub>before</sub> day-1, AIC day-8, DIC day-8, and IFC<sub>after</sub> day-1.

### Pressure Pain Threshold

The pressure pain threshold (PPT) was recorded by an investigator blind to the interference condition (PF) by means of an electronic algometer (Somedic) applied every day during IFC<sub>before</sub>; five times during the active or dummy interference conditions AIC or DIC, ie, at days 1, 2, 3, 5 and 8; and, again, every day after removal of the gold foil (IFC<sub>after</sub>).

**Table 1 Descriptive Statistics for the EMG Variables During Each Experimental Condition**

	Mean $\pm$ SD	5th percentile	95th percentile	Median	F ratio <sup>a</sup>	P	Post-hoc <sup>b</sup>		
							DIC	AIC	IFC <sub>after</sub>
<b>APs/h</b>									
IFC <sub>before</sub>	56.6 $\pm$ 19.9	8.8	94.0	48.9	0.1	.83	NS	NS	NS
DIC	53.4 $\pm$ 17.0	16.5	98.7	55.1			–	NS	NS
AIC	54.8 $\pm$ 24.7	2.9	108.5	48.2			–	–	NS
IFC <sub>after</sub>	57.9 $\pm$ 25.1	20.3	137.9	50.2					
<b>Dur (s)</b>									
IFC <sub>before</sub>	3.1 $\pm$ 2.3	0.5	12.0	1.0	0.5	.98	NS	NS	NS
DIC	3.0 $\pm$ 1.6	0.5	10.0	1.0			–	NS	NS
AIC	2.8 $\pm$ 1.2	0.5	10.0	1.0			–	–	NS
IFC <sub>after</sub>	2.9 $\pm$ 1.4	0.5	10.5	1.0					
<b>A<sub>mean</sub> (% MVC)</b>									
IFC <sub>before</sub>	17.2 $\pm$ 1.7	11.3	31.6	15.0	5.0	.01*	NS	NS	NS
DIC	18.0 $\pm$ 1.3	11.3	34.9	15.5			–	*	NS
AIC	15.9 $\pm$ 1.3	11.3	25.1	14.5			–	–	NS
IFC <sub>after</sub>	17.9 $\pm$ 1.8	11.3	35.0	15.2					

<sup>a</sup>Degrees of freedom = [3,18].

<sup>b</sup>Post-hoc multiple comparisons were Bonferroni corrected; level of significance: \* $P < .05$ ; \*\* $P < .01$ ; \*\*\* $P < .001$ .

EMG = electromyographic; Aps/h = activity periods per hour; Dur = net duration; A<sub>mean</sub> = mean amplitude; MVC = maximum voluntary contraction; IFC<sub>before</sub> = interference-free condition before gold foil application; IFC<sub>after</sub> = interference-free condition after gold foil application; DIC = dummy interference condition; AIC = active interference condition; NS = not significantly different.

The instrument and the procedure to record the PPT have been described in detail.<sup>19</sup> Briefly, the tip of the algometer had a surface of 1 cm<sup>2</sup>, and the rate of pressure increase was 20 kPa/s.<sup>30</sup> The PPT was determined as the point at which the pressure stimulus applied to the skin changed from a pressure sensation into a sensation of pain. PPT values were assessed bilaterally at four sites located on the masseter and anterior temporalis muscles, and at the thenar eminence. To ensure precise relocation of the face recording sites at each session, a transparent pliable plastic template was aligned to the ear, labial margin, and eye, and the location of the sites was marked. The sites were measured randomly with an interval of 5 seconds between sites. Four PPT measurements were made at each recording site, with a 2-minute rest interval between trials. In total, 10 minutes were needed to record PPTs at all sites. As the first PPT measurement within a session generally yields a higher value than that of the following measurements,<sup>31</sup> the value obtained from the first measurement was discarded and the PPT at each site was determined as the mean of the three subsequent measurements.

### Statistical Analyses

Normality of data was checked by the Kolmogorov-Smirnov test (K-S). The EMG data were analyzed by means of analysis of variance (ANOVA) for repeated measurements with post-hoc Bonferroni correction. Where appropriate, data were log converted. ANOVA was also used to analyze VAS ratings and PPT values

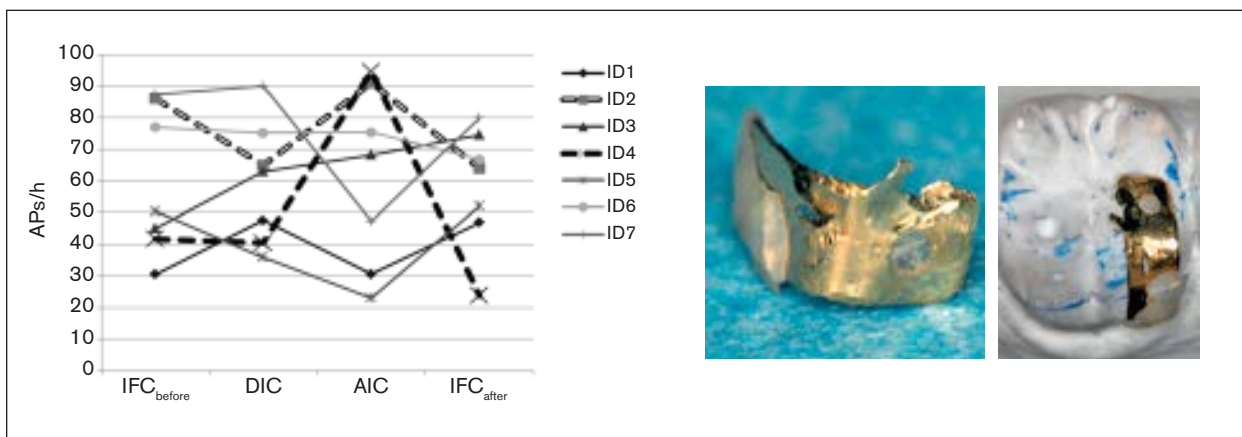
(SPSS 10.0, SPSS Inc). The significance level was set at  $P < .05$  (two-tailed).

## Results

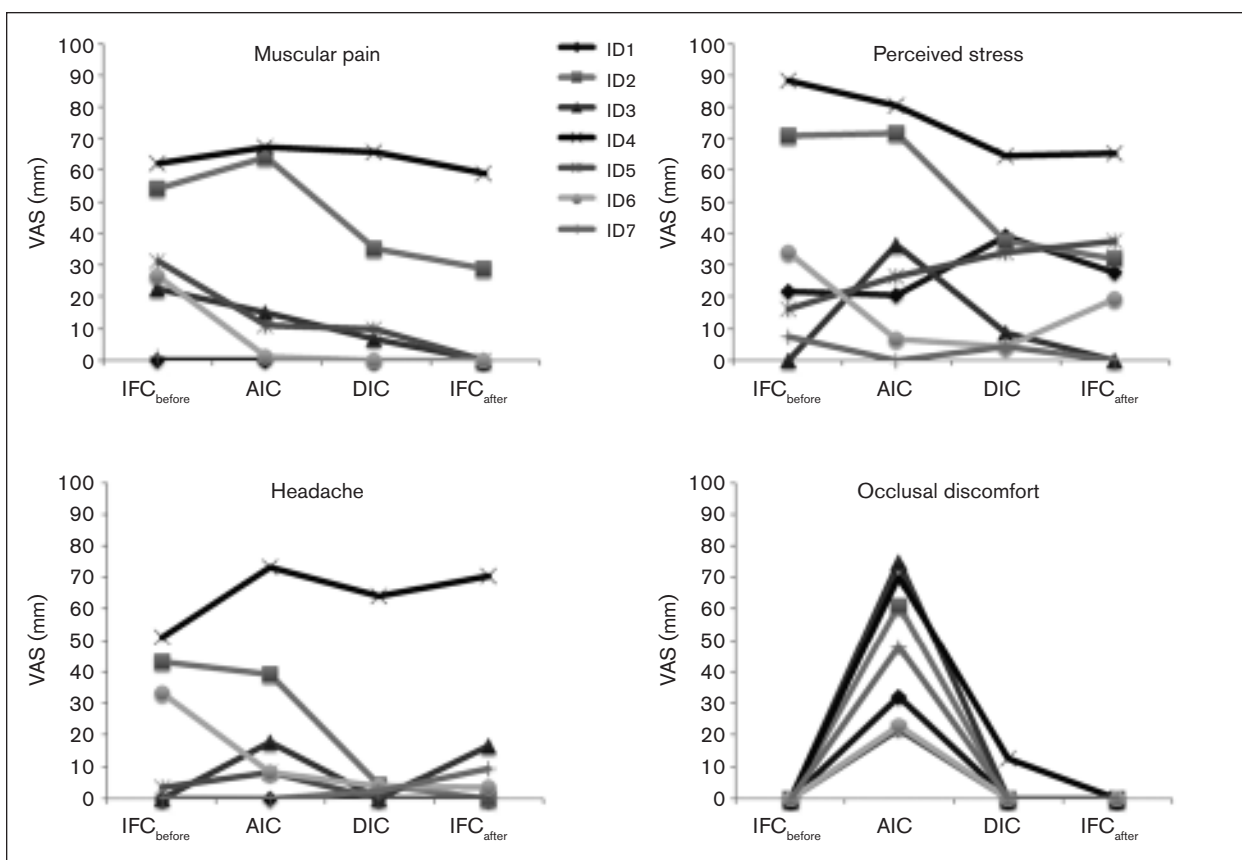
The study was carried out over a 10-month period. The time lag between the first TMD examination performed to recruit the study sample and the start of the experiment ranged between 1 and 8 months. One participant withdrew at day 1 of the AIC task due to lack of time to continue the experiment. Hence, the final sample included seven female volunteers (mean age  $\pm$  SD = 24.6  $\pm$  6.2 years).

At the initial screening examination, five participants reported TMD pain lasting for more than 6 months, one for 3 months and one for 1 month. Of the five whose pain lasted more than 6 months, three had a GCPS score of III (high disability, moderately limiting) and two had a GCPS score of II (low disability, high intensity). The participant with pain lasting 3 months had a GCPS score of I (low disability, low intensity), while the GCPS score of one participant was missing. The mean ( $\pm$  SD) score for facial pain intensity assessed by the numeric rating scale during the screening procedure (Question #7, RDC/TMD) was 5.8 ( $\pm$  0.4). According to the SCL-90-R, two female participants had severe and three moderate depression, and one was not depressed (one score missing).

The mean EMG amplitude of the APs, their frequency (APs/h), and the duration for each of the ex-



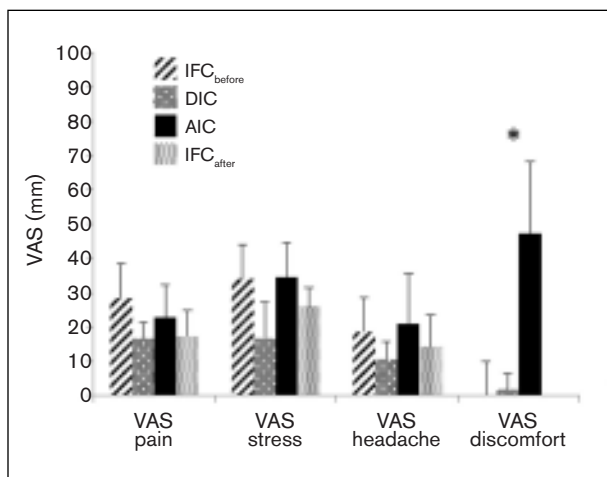
**Fig 2** Mean values of the activity periods per hour (APs/h) assessed during the four experimental sessions for each volunteer (ID1–ID7), and debonded gold foil of subject #2. Note the wear of the gold foil. Same abbreviations as in Fig 1. The data of only seven subjects are presented because one subject withdrew (see text).



**Fig 3** Mean VAS scores (mm) for muscular pain, perceived stress, headache, and occlusal discomfort during the four experimental conditions for each volunteer (ID1–ID7). Same abbreviations as in Fig 1.

perimental conditions are reported in Table 1 and Fig 2. The frequency and duration of the APs recorded during wakefulness did not differ significantly between the experimental conditions ( $P > .05$ ), but the mean EMG amplitude was significantly lower during the AIC than the DIC ( $P < .05$ ).

The individual VAS scores for facial pain, occlusal discomfort, headache, and perceived stress averaged for each condition are reported in Fig 3 and the corresponding group mean values ( $\pm$  SD) in Fig 4. The VAS scores for headache, facial pain, and stress did not differ significantly either between experimen-



**Fig 4** Group mean VAS scores (mm) for facial pain, perceived stress, headache, and occlusal discomfort during the experimental conditions. Bars indicate standard deviation. Same abbreviations as in Fig 1. \*Significant differences between experimental conditions.

**Table 2 Mean Pressure Pain Thresholds (PPT) for the Masseter and Temporalis Muscles Ipsilateral and Contralateral to the Occlusal Interference and for the Thenar Eminence**

Muscle site	Experimental condition	Mean PPT ± SD (KPa)
Masseter (ipsilateral)	IFC <sub>before</sub>	104.6 ± 86.8
	AIC	91.5 ± 23.8
	DIC	106.9 ± 47.6
	IFC <sub>after</sub>	114.8 ± 45.5
Masseter (contralateral)	IFC <sub>before</sub>	95.6 ± 36.3
	AIC	97.1 ± 25.5
	DIC	111.3 ± 43.1
	IFC <sub>after</sub>	116.5 ± 45.8
Temporalis (ipsilateral)	IFC <sub>before</sub>	103.1 ± 46.5
	AIC	96.1 ± 37.8
	DIC	112.7 ± 56.1
	IFC <sub>after</sub>	127.2 ± 60.5
Temporalis (contralateral)	IFC <sub>before</sub>	99.3 ± 46.7
	AIC	100.3 ± 34.0
	DIC	112.6 ± 57.2
	IFC <sub>after</sub>	124.3 ± 53.2
Thenar eminence	IFC <sub>before</sub>	175.4 ± 62.3
	AIC	178.0 ± 55.5
	DIC	184.0 ± 78.4
	IFC <sub>after</sub>	185.1 ± 60.9

tal conditions or between the different days within each condition ( $P > .05$ ). On the other hand, the occlusal discomfort increased significantly during the AIC ( $P < .001$ ).

The PPT values for masseter and temporalis muscles did not change significantly between the different experimental conditions at each measurement

site ( $P > .05$ , Table 2). TMD signs and symptoms, as assessed during the clinical examinations, did not change throughout the experimental conditions.

Table 3 compares the EMG data obtained in the present study with those obtained in an earlier sample of healthy TMD pain-free females receiving an active occlusal interference.<sup>11</sup>

## Discussion

This study investigated the effects of an occlusal interference in female volunteers affected by masticatory muscle pain during wakefulness. The results revealed that the application of an active occlusal interference had little influence on the contraction pattern of the masseter muscle. Indeed the frequency and duration of APs did not significantly change with the introduction of the active interference. Only the mean EMG amplitude of the APs decreased significantly during the active interference by about 2%, this reduction most likely being clinically irrelevant.

These results contrast with those of a previous study<sup>11</sup> that also recorded the APs during the same time windows and showed a marked reduction in the number and amplitude of the APs during wakefulness after the placement of an active interference in healthy TMD pain-free female participants (Table 3). The comparison with the previous findings yields another interesting observation: The female participants with myofascial pain had an approximately 20% higher frequency of APs per hour, as shown by the recording performed before interference insertion, and this value remained consistently higher throughout all study conditions.

The different reactions to the interference of these female participants with myofascial pain compared to healthy women might be related to the fact that masticatory muscle pain patients have the habit of holding the teeth in contact more often than healthy individuals during wakefulness.<sup>32-38</sup> Furthermore, masticatory muscle pain is more often diagnosed in individuals with a high frequency than a low frequency of oral parafunctions during wakefulness.<sup>39,40</sup> Hence, the avoidance behavior to the interference previously described in healthy female volunteers<sup>11</sup> seems to be lacking in subjects with myofascial pain. The lack of adaptation also supports the finding of Le Bell and coworkers, who reported that subjects with a history of TMD adapted less well to the introduction of an active occlusal interference than subjects without a TMD history.<sup>10,22</sup>

The present study was restricted to the effect of an occlusal interference during wakefulness because (1) wake-time parafunctions and sleep bruxism have different etiologies and (2) a previous study assessed

**Table 3 Descriptive Statistics for the EMG Variables During Each Experimental Condition for the TMD Group (Current Study) and for the TMD-Free Group (Michelotti et al<sup>11</sup>)**

	TMD group					TMD-free group <sup>a</sup>				
	Mean ± SD	<i>P</i>	DIC	AIC	IFC <sub>after</sub>	Mean ± SD	<i>P</i>	DIC	AIC	IFC <sub>after</sub>
<b>APs/h</b>										
IFC <sub>before</sub>	56.6 ± 19.9	.83	NS	NS	NS	48.0 ± 27.1	< .01	NS	**	NS
DIC	53.4 ± 17.0		–	NS	NS	44.4 ± 23.1		–	**	**
AIC	54.8 ± 24.7		–	–	NS	27.1 ± 18.7		–	–	*
IFC <sub>after</sub>	57.9 ± 25.1					42.2 ± 23.2				
<b>Dur (s)</b>										
IFC <sub>before</sub>	3.1 ± 2.3	.98	NS	NS	NS	2.7 ± 1.0	> .05	NS	NS	NS
DIC	3.0 ± 1.6		–	NS	NS	2.6 ± 0.9		–	NS	NS
AIC	2.8 ± 1.2		–	–	NS	3.0 ± 1.2		–	–	NS
IFC <sub>after</sub>	2.9 ± 1.4					2.4 ± 0.8				
<b>A<sub>mean</sub> (% MVC)</b>										
IFC <sub>before</sub>	17.2 ± 1.7	.01	NS	NS	NS	16.5 ± 1.8	< .001	NS	***	NS
DIC	18.0 ± 1.3		–	*	NS	16.2 ± 1.9		–	***	**
AIC	15.9 ± 1.3		–	–	NS	14.3 ± 1.0		–	–	**
IFC <sub>after</sub>	17.9 ± 1.8					16.2 ± 1.8				

<sup>a</sup>Data retrieved from Michelotti et al<sup>11</sup>. Post-hoc multiple comparisons for both the studies were Bonferroni corrected; level of significance: \**P* < .05; \*\**P* < .01; \*\*\**P* < .001; NS = not significantly different. Both the studies presented a similar research design. Between-group statistical comparisons were not computed because data belonged to different studies. See Table 1 for additional abbreviations.

the effect of occlusal interferences only on wake-time muscle activity.<sup>11</sup> All participants rated the occlusal discomfort provoked by the occlusal interference as very high. Moreover, the discomfort did not decrease during the active interference application time. This might be explained in part by the fact that the frequency of APs during the active interference time did not decrease. Indeed, in the previous study performed by this research group in TMD-free individuals, the decrease in occlusal discomfort was paralleled by a decrease in the frequency of medium- to high-intensity APs.<sup>11</sup> This explanation requires, of course, the assumption that during an AP the teeth were in contact. This assumption is legitimate, as the threshold level was set at 10% and a contraction of the masseter muscle of about 5% MVC is sufficient to bring the teeth in contact (see Table 1 in Roark et al).<sup>41</sup> The observation that the tooth pain induced by the interference did not cause a reduction of the number of APs fits well with empirical observations suggesting that habits are difficult to change. Indeed, habit suppression requires an active cognitive involvement from the patient, as is the case with habit-reversal therapy.<sup>42,43</sup>

The response to the active interference differed across individuals for both the frequency of APs and VAS pain scores. In particular, the two subjects with the greatest number of APs during the active interference period (subjects #2 and 4) also had the highest VAS pain scores. These participants had a low disability degree according to the GCPS (chronic pain grades II and I, respectively) but they had severe depression. One of these two subjects even debonded

the gold foil, which presented severe wear. However, she was not excluded from the study because the foil debonded on the last day of the AIC and was re-bonded within 1 hour.

The VAS scores for facial pain, headache, and perceived stress did not vary significantly between the experimental conditions, meaning that an occlusal interference does not affect pain intensity in female participants with myofascial TMD pain. The PPT values were within the ranges previously found for TMD participants, were lower than for healthy subjects,<sup>44,45</sup> and did not change significantly during the different conditions. This result, together with the EMG data and the VAS scores, further confirms that the interference did not significantly affect the behavior of the masseter muscle in these female participants. Even if they were extremely bothered by the occlusal interference, they maintained their usual masseter contraction pattern characterized by frequent contractions during wakefulness.

This study did have some limitations. First, the short duration of the experiment does not allow inferring whether a longer experimental window would have resulted in different results. However, the lack of significant differences in the frequency of activity periods between days, and therefore the lack of a trend, suggests that a longer time window may not have triggered a different response. Another limitation was the small size of the sample, which was primarily due to the difficulty in recruiting subjects willing to wear the electrodes during the day. The sample included only female student volunteers diagnosed with

myofascial pain and not patients seeking treatment. These two shortcomings limit the external validity of the study. It should also be noted that the EMG recordings obtained by means of portable recorders during wakefulness may be contaminated by artifacts due to movements of the electrodes. However, the distribution of these artifacts should be similar across the conditions and therefore should not have influenced the results. In addition, a threshold higher than the artifact noise was chosen. Furthermore, the EMG recordings by means of portable recorders do not allow distinguishing functional from nonfunctional masseter contractions. However, it has been shown that the number of functional tooth contacts does not differ between TMD patients and healthy subjects.<sup>32,46,47</sup> Moreover, despite the use of a template to reposition the electrodes in the same location, it is not certain that the activity of the same muscle fibers was recorded throughout the entire experiment. Unfortunately, it is impossible to know whether this problem, which is inherent to long-time surface EMG recording approaches, affected the results. Finally, this study design did not provide information about the effect of an occlusal interference on sleep bruxism, which is a different entity than wake-time parafunctions, and was not able to provide information about possible central and peripheral mechanisms regulating the activity of the masticatory muscles under the experimental interference condition; some of these mechanisms, however, have been addressed in animal models.<sup>17,18</sup>

## Conclusions

Female volunteers with masticatory muscle pain feel particularly hampered by the introduction of an experimental occlusal interference. Nevertheless, they do not show an avoidance behavior, since they did not significantly change the pattern of habitual masseter EMG activity during wakefulness. This contrasts the avoidance behavior recorded in pain-free female volunteers, who demonstrated a reduction of the masseter EMG activity after insertion of an active interference.

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