Female Hormonal Fluctuation and Masticatory Function in Patients with Disc Displacement—A Case-Control Study

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Purpose: The aim of this case-control study was to investigate the role female hormone fluctuation plays in maximum occlusal force and masticatory performance in female subjects presenting disc displacement with reduction (DDR) and matched controls. ***Materials and Methods:*** Sixty-five subjects were initially recruited; however, 4 of them were excluded during the study. The final sample included 14 subjects with DDR taking oral contraceptives, 16 control subjects without DDR taking oral contraceptives, 14 normally cycling subjects with DDR, and 17 normally cycling subjects without DDR. DDR was diagnosed by means of the Research Diagnostic Criteria for Temporomandibular Disorders axis I, and subjects without pain were selected. Maximum occlusal force was measured bilaterally in the molar region using a force transducer, and masticatory performance was analyzed using the artificial material comminution and sieving method. Variables were evaluated in four phases of three menstrual cycles, identified by ovulation testing. Data were submitted to the Mauchly sphericity test and PROC MIXED procedure of the SAS statistical program for repeated measures. Multiple comparisons were made using the Tukey-Kramer test ($P \leq .05$). ***Results:*** Comparisons among menstrual cycle phases showed no differences in occlusal force ($P = .44$) or masticatory performance ($P = .09$) for all volunteers. Subjects without DDR showed greater occlusal force ($P = .01$). No difference in occlusal force was found between subjects regardless of whether they took contraceptives ($P = .15$). Similarly, masticatory performance values did not differ among subjects with or without DDR ($P = .09$) or among those taking or not taking contraceptives ($P = .29$). ***Conclusion:*** Hormonal fluctuation did not influence mastication. *Int J Prosthodont 2011;24:320–327.*

Temporomandibular disorder (TMD) is a collective term that comprises a number of clinical conditions involving the masticatory muscles or temporomandibular joints and associated structures.[1] The multifactorial etiology of TMD[2,3] could involve acute trauma,[1] occlusal aspects,[1,3] parafunctional habits,[1] and psychologic,[4] emotional, or psychosocial factors.[5]

It is suggested that females are more likely to develop and maintain TMD.[6] Furthermore, the disturbances are more prevalent in female subjects of reproductive age, suggesting a possible relationship between TMD and normal hormonal fluctuation during the menstrual cycle,[7] characterized by continuous changes in four hormones: estrogen, progesterone, follicle stimulating hormone, and luteinizing hormone.[8] Estrogen is considered to be involved in the pathogenesis of TMD,[6] and several studies have provided evidence for the hypothesis that estrogen may influence the development, restitution, and metabolism of the temporomandibular joints and associated structures.[6,9,10]

Hormonal fluctuation may also be responsible for the onset or increase in physical symptoms (eg, headache), psychiatric disorders (eg, depression and somatization),[4] and emotional alterations (eg, stress) that may affect the tonus of the masticatory muscles, increasing their activity,[11] the load on the teeth, and altering occlusal force.

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Occlusal force contributes to masticatory function and aids in the process of comminuting food, triturating it in preparation for swallowing,[12] and is considered an important tool for evaluating masticatory function.[13] Maximum occlusal force magnitude depends on several variables, such as the number and extension of occlusal contacts,[14] sex,[13,15] and presence of signs and symptoms of TMD.[16,17]

Masticatory function can also be analyzed by masticatory performance, which can be defined as the food particle size distribution after a given number of chewing strokes.[18] Masticatory function could be influenced by the number of occlusal pairs,[15] occlusal force,[15] the presence of removable prostheses, and signs and symptoms of TMD.[19,20]

The literature is inconclusive regarding the influence of TMD on masticatory function. The majority of studies[17,19,20] have indicated that TMD patients have lower masticatory capacity in comparison with matched controls, especially when pain symptoms are present; however, there is no consensus.[21] Considering that masticatory function could be altered by the presence of TMD[19,20] and that estrogen could play a role in the maintenance of TMD by the interplay with collagen metabolism,[9] it could be hypothesized that menstrual cycle–related hormonal fluctuation could influence masticatory function. Studies have been conducted on hormonal fluctuation and its effects on the temporomandibular joint of TMD patients[2,22,23]; however, no reports were found on the influence of female hormones on maximum occlusal force and masticatory performance. Thus, the aim of this study was to investigate the role of sex hormone fluctuation in the maximum occlusal force and masticatory performance of females diagnosed with nonsymptomatic disc displacement with reduction (DDR) and matched controls.

Materials and Methods

A convenience sample initially composed of sixty-five dentate female subjects (age range: 17 to 43 years) was selected from among students and staff of the Piracicaba Dental School, State University of Campinas, São Paulo, Brazil, to participate in this study. The inclusion criteria were to present with: *(1)* good general health, with no systemic or psychologic diseases, and not taking any medication, except for oral contraceptives (OCs); *(2)* good oral health with no caries or periodontal disease; *(3)* complete dentition (except for missing third molars); *(4)* no composite or amalgam restorations in the first molars; *(5)* no malocclusion (anterior open bite, unilateral or bilateral posterior crossbite); *(6)* no facial deformities (facial asymmetry, cleft palate, cleft lip, masseteric hypertrophy); and *(7)* no parafunctional habits, such as bruxism or tooth grinding, diagnosed through clinical examination of occlusal wear and a patient's self-report of tooth grinding.[24] Normally cycling subjects had to have regular menstrual cycles varying between 24 and 32 days and must not have used OCs for at least 3 months. Subjects using OCs had to be using a low-dose combination of exogenous sex hormone (estrogen and progesterone) pills in a 21-day cycle and had to have been on the pill regimen for at least 3 months. Pregnant or menopausal subjects and those with hormonal disease, undergoing fertility treatment, with TMD pain symptoms, and those wearing any type of dental prosthesis were excluded from the study. After selection, three subjects were excluded (one became pregnant and two changed their OC dosage). The research protocol was approved by the ethics committee of the Piracicaba Dental School, and a signed consent form was obtained from all participants.

At baseline, all subjects were submitted to the axis I of the Research Diagnostic Criteria for Temporomandibular Disorders (RDC/TMD).[25] Those who were selected and presented with DDR comprised the experimental group. At that time, 1 subject presented disc displacement without reduction and was excluded from the research. Thus, the final sample was composed of 61 subjects. Controls were selected according to the same criteria, except that they could not present DDR. After this, the recruited subjects were divided according to the presence of DDR and the intake of OCs as follows: 14 subjects with DDR taking OCs (mean age: 23.21 ± 3.01 years), 16 control subjects without DDR taking OCs (mean age: 22.8 ± 3.17 years), 14 normally cycling subjects with DDR (mean age: 24.43 ± 5.97 years), and 17 normally cycling subjects without DDR (mean age: 23.9 ± 5.7 years). The number of subjects in each group was determined by preliminary tests, which demonstrated that the sample size yielded an adequate power (0.8) for detecting statistically significance differences.

In the same appointment, all volunteers underwent anthropometric measurements. They were measured and weighed on a digital scale (PL200, Filizola) barefoot, standing with their feet together and with the Frankfort plane parallel to the floor. Body Mass Index was calculated as the ratio between weight in kilograms and height in meters squared (kg/m^2).

The menstrual cycle was divided into four phases,[26] considering the last menses, ovulation period, and the length of the cycle as follows: *(1)* menstrual: from the first day of bleeding to the fifth day; *(2)* follicular: from the sixth to eleventh day; *(3)* ovulatory: from the twelfth to sixteenth day, determined by means of the BioEasy

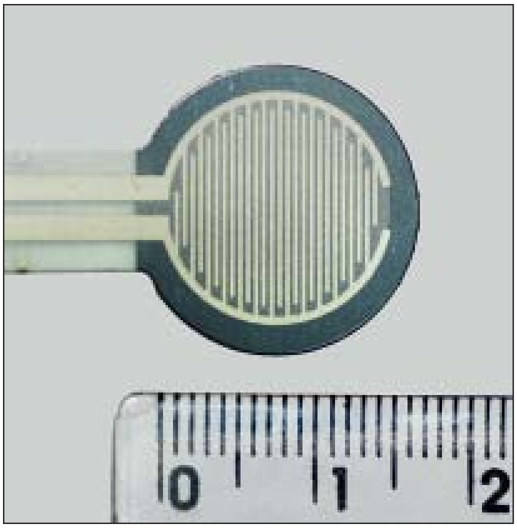

Fig 1 *(left)* Sensor used for maximum occlusal force evaluation.

Fig 2 *(below)* Maximum occlusal force sensors positioned in the first molar region.

ovulation prediction test (BioEasy Diagnóstica); and *(4)* luteal: between the twenty-fourth and twenty-eighth day of the menstrual cycle. The menstrual phase was determined by self-report on the first day of bleeding, when subjects were instructed to call the researcher. At the expected time of ovulation, women who were not taking OCs were called by the researcher and instructed on how to use the ovulation prediction test to determine the ovulatory phase.

Maximum occlusal force and masticatory performance were evaluated for three complete menstrual cycles, and at each evaluation time, the researcher was blinded to the RDC/TMD data. In case of a non-ovulatory cycle, measurements were carried out in the following ovulatory menstrual cycle. All evaluations were performed by a single researcher in the four phases of the menstrual cycle, scheduled for approximately the same time of day. To determine the data variability, the maximum occlusal force and masticatory performance of subjects taking OCs were also evaluated 4 times a month over a period of 3 months. Thus, all volunteers were evaluated for 3 complete months, yielding a total of 12 appointments.

Maximum Occlusal Force

Bilateral maximum occlusal force was evaluated using an occlusal force transducer (Spider 8, Hottinger Baldwin Messtechnik) composed of two sensors (FSR no. 151, Interlink Electronics).[27] The validation and reproducibility of these sensors were verified by Fernandes et al.[27] The signals of each sensor were recorded, amplified, and analyzed using the software Catman Easy 1.0 (Hottinger Baldwin Messtechnik). The sensors (Fig 1) were 1.2 mm in diameter and 0.25-mm thick and were protected from deformities during clenching on both sides by 1.0-mm-thick metal disks of the same diameter and rubber disks 1.7-mm wide, resulting in a 5.65-mm-wide assembly. The operator placed these sensors bilaterally in the first molar regions, and subjects were requested to occlude with maximum force for 7 seconds (Fig 2), measured by the equipment software. A rest of 5 minutes was allowed, and the procedure was repeated. The highest measurement value as a result of the maximum occlusal force was used and recorded in kgf.

Reproducibility of maximum occlusal force measurements was verified by using intraclass correlation coefficients (ICCs), assessed from the two measurements of each appointment during the 3 months for all volunteers at a confidence interval of 95%. The ICC was 0.92 for the first month, 0.93 for the second, and 0.94 for the third month, which was considered excellent.[28]

Masticatory Performance

Masticatory performance was evaluated using the sieve method. Subjects were instructed to chew a portion of 17 cubes with an edge size of 5.6 mm (3 cm^3) composed of an artificial silicon test material (Optosil, Heraeus Kulzer) in their habitual way for 20 chewing strokes,[18,29] counted by the operator. The particles obtained were expectorated onto a paper

Table 1 Sample Characteristics (Mean ± Standard Deviation)

	With DDR taking OCs (n = 14)	Without DDR taking OCs (n = 16)	With DDR not taking OCs (n = 14)	Without DDR not taking OCs (n = 17)	P*
Age (y)	23.21 ± 3.01	22.80 ± 3.17	24.43 ± 5.97	23.90 ± 5.70	.92
BMI (kg/m^2)	21.82 ± 2.53	21.26 ± 2.20	21.49 ± 4.07	21.97 ± 2.60	.89

BMI = Body Mass Index.
*One-way analysis of variance.

filter placed on a glass container. Mouth rinses were performed with 200 mL of water and then expectorated onto the same filter several times to completely cleanse the oral cavity. Finally, the subjects' mouths were examined for pieces of retained test material. After this, the water was completely drained, and the filter with the particles was stored in an electric oven at 80°C for 25 minutes.[30] A sieving machine (Bertel Industria Metalurgica) was used for 20 minutes to sieve the particles through a stack of up to 10 sieves, with mesh sizes gradually decreasing from 5.6 to 0.5 mm, and a bottom plate. The amount of test material retained on each sieve and on the bottom plate was weighed on an analytic balance reading to 0.001 g (Model 2060, Bel Engineering).[18,29] Masticatory performance was calculated as the median particle size (X_{50}), which is the aperture of a theoretic sieve through which 50% of the weight of the comminuted food could pass.[18,31] The cumulative distribution of the particle sizes by weight can be described mathematically by the Rosin-Rammler equation:

$$Q_w^-(X) = 1 - 2^{(-X/X_{50})^b}$$

where Q_w is the weight fraction of the particles with a size smaller than X, and b represents the size spread of the distribution (broadness variable).[31] The validation and reproducibility of this method was reported by van der Bilt et al.[31]

Statistical Analysis

Sample homogeneity according to age and Body Mass Index were analyzed by one-way analysis of variance. The Mauchly sphericity test was applied, and the sphericity was violated. Therefore, the PROC MIXED procedure of the SAS statistical program (release 9.1, SAS Institute) was applied for repeated measures. Multiple comparisons among groups and menstrual cycle phases were made by the Tukey-Kramer test. The Pearson correlation coefficient was calculated between the results for maximum occlusal force and masticatory performance. All tests were carried out at a significance level of 5%.

Results

Sample Characteristics

Table 1 shows the summary of the anthropometric characteristics of the sample. Homogenous distribution was observed among groups ($P > .05$). There was no statistical difference among groups for age ($P = .92$) or Body Mass Index ($P = .89$).

Maximum Occlusal Force

In a general analysis, irrespective of hormonal fluctuation, DDR subjects presented decreased maximum occlusal force values when compared with matched controls ($P = .01$) (Table 2). There was no significant difference in maximum occlusal force values between subjects whether or not they took OCs ($P = .15$). Comparisons among menstrual cycle phases for normally cycling subjects and among evaluations for OC users also showed no difference in maximum occlusal force values for all groups studied ($P = .44$).

Masticatory Performance

Irrespective of hormonal fluctuation, DDR subjects and matched controls showed no differences in masticatory performance ($P = .09$) (Table 3). No differences in masticatory performance were noted among menstrual cycle phases for normally cycling subjects, among evaluation times for subjects taking OCs ($P = .09$), or between OC groups ($P = .29$) (Table 3).

The Pearson correlation coefficient between maximum occlusal force and X_{50} values was $r = -0.3238$ (95% confidence interval: −0.39 to −0.26, $P < .0001$).

Discussion

In this study, an endeavor was made to elucidate whether female hormone fluctuations had an influence on maximum occlusal force and masticatory performance. There are several studies concerning TMD pain and the phases of the menstrual cycle.[22,23,26,32]

Table 2 Mean Values for Maximum Occlusal Force (kgf) According to Presence of DDR, Use of OCs, and Evaluation Period

	DDR	
OC/Evaluation period	With (SE, SD)	Without (SE, SD)
Taking		
First assessment (bleeding)	54.80 (± 3.14, ± 14.60)	62.16 (± 2.94, ± 15.82)
Second assessment	53.70 (± 3.14, ± 10.97)	59.50 (± 2.94, ± 13.36)
Third assessment	55.23 (± 3.14, ± 11.94)	65.19 (± 2.94, ± 11.96)
Fourth assessment	54.18 (± 3.14, ± 16.08)	64.54 (± 2.94, ± 10.85)
Not taking		
Menstrual phase	51.64 (± 3.20, ± 7.25)	56.71 (± 3.58, ± 10.27)
Follicular phase	52.43 (± 3.20, ± 9.37)	58.15 (± 3.58, ± 10.69)
Ovulatory phase	51.26 (± 3.20, ± 5.25)	58.83 (± 3.58, ± 13.85)
Luteal phase	51.61 (± 3.20, ± 8.11)	56.62 (± 3.58, ± 12.09)
Mean	53.10 (± 1.95, ± 10.45)[a]	60.21 (± 1.78, ± 12.33)[b]

SE = standard error; SD = standard deviation.
Means followed by different letters are statistically different ($P \leq .05$).

Table 3 Mean Values for Masticatory Performance (X_{50}) According to Presence of DDR, Use of OCs, and Evaluation Period

	DDR	
OC/Evaluation period	With (SE, SD)	Without (SE, SD)
Taking		
First assessment (bleeding)	4.84 (± 0.20, ± 0.86)	4.26 (± 0.19, ± 0.63)
Second assessment	4.80 (± 0.20, ± 0.66)	4.23 (± 0.19, ± 0.54)
Third assessment	4.80 (± 0.20, ± 0.75)	4.10 (± 0.19, ± 0.55)
Fourth assessment	4.83 (± 0.20, ± 0.74)	4.11 (± 0.19, ± 0.59)
Not taking		
Menstrual phase	4.68 (± 0.20, ± 0.74)	4.66 (± 0.18, ± 0.82)
Follicular phase	4.77 (± 0.20, ± 0.74)	4.80 (± 0.18, ± 0.87)
Ovulatory phase	4.71 (± 0.20, ± 0.87)	4.68 (± 0.18, ± 0.85)
Luteal phase	4.62 (± 0.20, ± 0.71)	4.66 (± 0.18, ± 0.85)
Mean	4.75 (± 0.20, ± 0.76)[a]	4.44 (± 0.13, ± 0.71)[a]

SE = standard error; SD = standard deviation.
Means followed by the same letter are not statistically different ($P > .05$).

However, internal derangements such as DDR can persist for a long period of time without the presence of pain,[1,21] and little is known about the effects of hormones on pain-free patients. Considering that maximum muscle output during forceful muscle work is reduced in the presence of pain,[33] maximum occlusal force and masticatory performance would also be influenced by painful symptoms. Thus, subjects in pain were excluded from the study to avoid confounding variables, which could mask the relationship between the objective variables of mastication and hormone variations.

In addition to the pain-free condition, the study sample was composed of students and staff from a dental school, which could be considered a potential bias factor.[28] However, maximum occlusal force and masticatory performance measurements were not influenced by previous awareness because to minimize bias, care was taken to select first-year undergraduate students and employees working in the offices who are not supposed to have knowledge on the topic. Regarding sample size, it should be emphasized that there was low data variability for both maximum occlusal force and masticatory performance variables, which revealed that there was a sufficient number of subjects in each group for statistical analysis. It should also be considered that in spite of the low variability of the data for maximum occlusal force, no significant differences were observed between groups taking or not taking OCs or among menstrual cycle phases because the difference was very small and, therefore, not clinically relevant. Nevertheless, for groups with and without DDR, the difference was important and statistically significant. For masticatory performance, no significant differences were observed in spite of the low variability and the large number of volunteers, a result of these differences being very small. The study was also standardized by age and Body Mass Index to improve the confidence level.

The main finding was that the variations occurring in reproductive hormones during the menstrual cycle did not influence maximum occlusal force and masticatory performance of patients with or without DDR. Some authors have reported the relationship between muscle strength and masticatory function,[15] and although few studies have examined the end result of the influence of the menstrual cycle on the muscular strength of masticatory muscles, the results of this study are consistent with findings on other muscles and synovial joints in the human body.[34] A study using electric stimulation to ensure maximum muscle contraction found no significant changes in quadricep muscle strength in female subjects throughout the phases of the menstrual cycle, nor did they find significant correlations between any of the strength indices and female reproductive hormone concentrations.[8] Moreover, according to Constantini et al,[34] muscular strength does not appear to fluctuate significantly during an ovulatory menstrual cycle. Thus, considering that maximum occlusal force and masticatory performance are related to muscle strength, the findings of the present study appear to be in agreement with those of the aforementioned authors.

Another mechanism that has been suggested as possibly being affected by female hormones is ligament laxity.[34] The change in laxity throughout the menstrual cycle may be a direct action of estrogen on the ligament,[34] leading to an increase in type I-trimer collagen, which is more susceptible to distention than normal collagen and is related to temporomandibular joint mobility and disc displacement.[9] Nevertheless, although estrogen receptors are present in some of the human body joints, such as in the anterior cruciate ligament of the knees[35] and the temporomandibular joints,[22] some authors have reported no significant differences in anterior cruciate ligament laxity among the four phases of the menstrual cycle.[36,37] Thus, if hormone fluctuation has no effect on ligament laxity, hormones may not have any effect on masticatory functions related to these ligaments, such as maximum occlusal force and masticatory performance,[19] even in DDR patients. Furthermore, studies concerning muscle strength[8,34] have also found no significant changes in muscle performance across the menstrual cycle, suggesting that variations in female hormones do not affect muscle contractile characteristics.

It has been reported that maximum occlusal force is not influenced by TMD.[13,38,39] Nevertheless, the findings of the present study showed that DDR subjects without pain presented reduced occlusal force values. Although there are few studies that have examined occlusal force in DDR subjects, the results of the present investigation are in agreement with those found by Sato et al[19] and Sinn et al,[40] who observed reduced occlusal force values in subjects presenting internal derangement of the temporomandibular joint, although the latter authors did not mention the pain condition of their subjects. According to Sato et al,[41] chewing movement is impaired in patients with anterior disc displacement, which could decrease the lateral mandibular movements generating masticatory muscle imbalance. Consequently, since maximum occlusal force is measured during maximal masticatory muscle contraction,[15] any change or imbalance in these muscles or in the temporomandibular joint would probably modify muscle activity and alter maximum occlusal force. Moreover, since maximum occlusal force was measured with the use of thin sensors, the articular disc was probably displaced at the time of recording the values, representing a mechanical interference in the mandibular closing movement and contributing to the results.

Although DDR subjects presented with decreased maximum occlusal force, they showed similar masticatory performance values when compared with matched controls. There are a few studies in the literature reporting on mastication and pain-free TMD subjects. However, the results of this study are consistent with those found by Ikebe et al,[21] who verified no difference in masticatory performance between asymptomatic subjects with and without temporomandibular joint sounds, although they studied an older Japanese population. On the other hand, the results of the present study are in contrast with those found by Sato et al[19] and Peroz and Tai,[20] who showed that the values of masticatory efficiency and masticatory performance, respectively, were lower in patients with nonreducing disc displacement of the TMJ. However, different from the present study, the former used a distinct methodology to verify mastication, which was measured by chewing adenosine triphosphate granules and calculated by the sample absorbance. The second study mentioned included patients with pain, which could explain the apparent discrepancies among these studies.

In spite of maximum occlusal force values having shown a difference between DDR subjects and controls, masticatory performance values were similar. This was not expected, since there has been a reported correlation between maximum occlusal force and masticatory performance,[15] and this was confirmed in the present study ($r = -0.3238$). When considering the asymptomatic nature of DDR subjects selected for this study and that the masticatory performance test does not require a maximum muscle effort to chew artificial test material[42] when compared with the occlusal force test, both experimental and control subjects could probably exhibit similar masticatory performance values, thus offering some support for this finding. In addition, considering that all subjects in the present study were completely dentate without malocclusion, one supposes that the number of occlusal contacts could contribute to explaining the masticatory performance values.

Although there was low individual variability, maximum occlusal force values obtained for control subjects were similar to those found previously.[15,17] However, they were lower than the values found by Sato et al[19] and Rudy et al.[43] The reasons may be related to methodologic differences. Masticatory performance values for control subjects were consistent with those obtained by Lujan-Climent et al.[44] Nevertheless, they were higher than those of Fontijn-Tekamp et al[18] and Pereira et al.[17] These disagreements may be a result of the differences in sample selection among reports, since the present study recruited only female subjects.

Over the course of an ovulatory cycle, there are predictable and measured variations in the female sex steroids that affect different body systems. However, there is no conclusive evidence of their effects on the masticatory system. Thus, there is great need for further studies on the objective functions of mastication in symptomatic TMD patients.

Conclusions

Within the limitations of the present study, it was possible to conclude that female hormonal fluctuation did not influence masticatory function. The presence of DDR was able to impair maximum occlusal force; however, it had no influence on masticatory performance.

Acknowledgments

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