Effects of therapy on masseter activity and chewing kinematics in patients with unilateral posterior crossbite

Maria Grazia Piancino[a,*], Deborah Falla[b], Andrea Merlo[c], Teresa Vallelonga[a], Corrado de Biase[a], Domenico Dalessandri[d], Cesare Debernardi[a]

[a] CIR Dental School, Department of Surgical Sciences, Division of Orthodontics, University of Turin, Italy
[b] Department of Neurorehabilitation Engineering, University Hospital Gottingen, Germany
[c] Motion Analysis Laboratory, Local Health Unit (AUSL) of Reggio Emilia, Department of Rehabilitation, Correggio, Italy
[d] School of Dentistry, Division of Orthodontics, University of Brescia, Italy

ARTICLE INFO

ABSTRACT

Objective: To describe the effects of therapy on masseter activity and chewing kinematic in patients with unilateral posterior crossbite (UPC).

Design: Fifty children (age: mean ± SD: 9.1 ± 2.3 years) with UPC (34 on the right side, 16 on the left side) and twenty children (age: 9.5 ± 2.6 years) with normal occlusion were selected for the study. The mandibular motion and the muscular activity during chewing soft and hard boli were simultaneously recorded, before and after correction with function generating bite, after a mean treatment time of 7.3 ± 2.4 months plus the retention time of 5–6 months. The percentage of reverse cycles and the percent difference between ipsilateral and contralateral peaks of the masseter electromyography envelopes were computed.

Results: Before therapy, the percentage of reverse cycles during chewing on the crossbite side was greater in patients than in controls (P < 0.001) and significantly reduced after therapy (P < 0.001) towards the reference normal value (soft bolus; pre: 57 ± 30%, post:12 ± 17%; hard bolus; pre: 65 ± 34%, post: 12 ± 13%; reference value: soft bolus 4 ± 2%, hard bolus 5 ± 3%). Before therapy the percent difference between electromyography envelope peaks in patients was lower than in controls (P < 0.01) and significantly increased after therapy (P < 0.05) becoming similar to the reference normal value.

Conclusions: The correction induced a normal-like coordination of masseter muscles activity together with a significant reduction of the reverse chewing patterns. The previous altered muscular activation corresponded to the altered kinematics of reverse chewing cycles that might be considered a useful indicator of the severity of the masticatory function involvement.

1. Introduction

Posterior unilateral crossbite is a serious asymmetric malocclusion, involving teeth, structures and functions of the stomatognathic system. It may become clinically evident at a very early stage in development, between 18 months and 5 years of age, during the eruption of the primary dentition, but it can also involve the permanent dentition at a later stage of development (da Silva Filho, Santamaria, & Capelozza Filho, 2007; Proffit, Fields, & Moray, 1998; Thilander & Lennartsson, 2002). Thus, when posterior unilateral crossbite affects children in early childhood it involves the components of the stomatognathic system that are actively developing, including motor control of masticatory function (Kiliaridis, Mahboubi, Raadsheer, & Katsaros, 2007; Nerder, Bakke, & Solow, 1999; Pinto, Buschang, Throckmorton, & Chen, 2001; Pirttiniemi, Kantomaa, & Lahtela, 1990; Thilander & Bjerklin, 2012).

Children with a unilateral posterior crossbite exhibit different types of unusual chewing patterns when chewing on the affected side, which were documented for the first time in the sixties (Ahlgren, 1967). The significant presence of reverse sequence chewing cycles, which refers to movement of the mandible during the closing phase of chewing as described later by Lewin and Ramadori (1985), has been well established in patients with

crossbite, with high occurrences observed during chewing on the crossbite side only (Ben-Bassat, Yaffe, Brin, Freeman, & Ehrlich, 1993; Lewin & Ramadori, 1985; Piancino et al., 2006; Piancino, Frongia, Dalessandri, Bracco, & Ramieri, 2013; Rilo, Silva, Mora, Cadarso-Suárez, & Santana, 2007; Throckmorton, Buschang, Hayasaki, & Pinto, 2001; Wilding & Lewin, 1994). Reverse chewing cycles show an abnormal, narrow kinematic pattern in the frontal plane, characterized by cross over of the tracings and limited lateral displacement of the mandible in comparison with the pattern of the non-affected side, which shows physiological morphology. As a result there is a serious asymmetry of the masticatory function (Lewin & Ramadori, 1985; Piancino et al., 2006; Sever, Marion, & Ovsenik, 2011).

Mirroring the kinematic pattern, the activation of the masseter muscles is altered and subjects with unilateral posterior crossbite show marked differences between sides resulting in an asymmetrical activation of the masseter muscles during rest, maximal clenching, and swallowing (Andrade, Gavião, Gameiro, & De Rossi, 2010; Ferrario, Sforza, & Serrao, 1999; Martín, Palma, Alamán, Lopez-Quiñones, & Alarcón, 2012; Piancino, Farina, Talpone, Merlo, & Bracco, 2009). Moreover during unilateral chewing on the side of crossbite, children with unilateral posterior crossbite show decreased activity of the masseter on the crossbite side and increased masseter activation on the contralateral side during reverse cycles (Piancino et al., 2009). This results in a reduced side to side difference in masseter muscle activity in children with unilateral posterior crossbite, whereas normally, unilateral chewing is characterized by a significant difference in activation between the ipsilateral and contralateral masseter muscle (Piancino, Bracco, Vallelonga, Merlo, & Farina, 2008).

It remains unknown whether functional therapy, which has been shown to correct reverse chewing cycles(Piancino et al., 2006), can re-establish the normal coordination between the bilateral masseter muscles during chewing. That is, whether functional therapy can induce an increase in activation of the ipsilateral masseter or decrease in contralateral masseter activity so that the difference in bilateral masseter muscle activation resembles that of control subjects. This knowledge is important to appreciate whether repositioning of the teeth within the dental arches can improve neuromuscular control of chewing (Johnsen & Trulsson, 2005; Lund, Scott, Kolta, & Westberg, 1999; Morquette et al., 2012; Quintero et al., 2013; Woda et al., 2010).

This study describes the effect of the functional therapy delivered at our center on both kinematic chewing patterns and masseter muscle coordination in patients with unilateral posterior crossbite.

2. Materials and methods

Fifty children (age: mean ± SD: 9.1 ± 2.3 years) with unilateral posterior crossbite (34 on the right side, 16 on the left side) and twenty children (age: 9.5 ± 2.6 years) with normal occlusion, referring to the Orthodontic Department of the University of Turin, Italy, from January 2011 through April 2014, were selected for this observational study. Before participating in the study, informed consent was obtained from the parents and the study was approved by the Institutional Review Board of the University Hospital "Health and Science Complex Turin-Italy" n. CS/246, in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans.

The inclusion criteria for the patients group were: (i) unilateral posterior crossbite of two or more posterior teeth, (ii) mixed dentition. The exclusion criteria were the presence of (i) previous orthodontic therapy, (ii) erupting teeth, (iii) caries, (iv) dental pain.

A parallel control group was carefully selected for normal occlusion and mixed dentition, and was matched with the patient group for age and gender.

Casts in maximal intercuspation and orthopantomography were obtained from all subjects and patients to classify, respectively, the type of crossbite and the erupting teeth.

The recordings of chewing cycles were carried out immediately before the intervention and after correction plus the retention time, whereas the control group was measured twice with a time span of six months. Both cases and controls data were analyzed in the same time period.

2.1. Appliance

Each patient was treated with the functional appliance 'Function Generating Bite' (Fig. 1). The appliances were individually manufactured and made of acrylic resin and special resilient stainless steel, with posterior metallic bite planes preventing the teeth from intercuspal contacts. The appliance is characterized by a
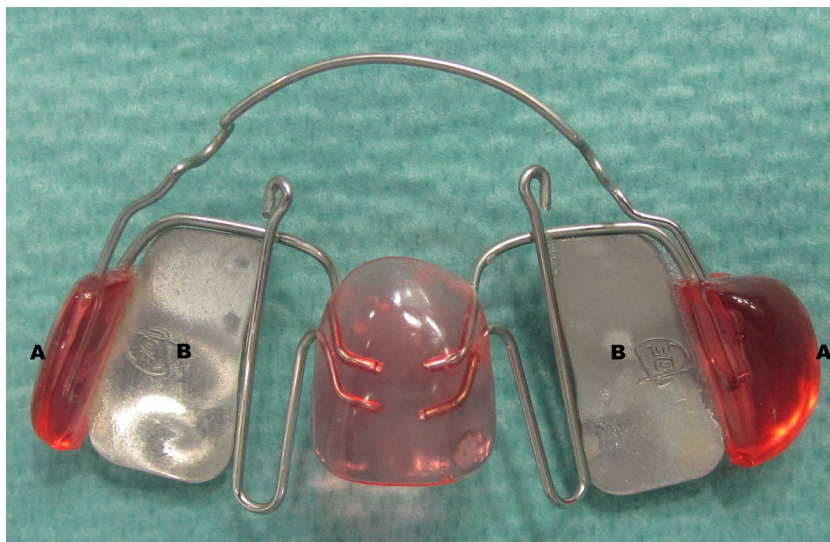
Fig. 1. The Function Generating Bite appliance, upper view. The resilience of the bite planes (B) and the elasticity of the wires of the appliance permit the orthodontic movement of the teeth avoiding dental trauma. Due to the muscular anchorage the different thickness of the buccal shields (A) lets the application of different forces between sides to correct the asymmetric malocclusion.

muscular anchorage, dental point contacts and intermittent forces. It is activated during swallowing. The teeth cusps compress the posterior metallic bite planes leveling the occlusal plane simultaneously to the action of the expansion springs. The resilience of the bites and the elasticity of the wires permit a gradual orthodontic movement avoiding dental trauma. The efficacy of the treatment has been addressed elsewhere (Piancino et al., 2006). At the end of treatment, the malocclusion resulted corrected and the buccal cusps of the upper teeth, which were previously in crossbite, overlapped the lower teeth on the buccal side, thus providing the appropriate physiological stimuli from peripheral receptors and proprioceptors (Piancino et al., 2006). The mean treatment time was 7.3 ± 2.4 months plus the retention time of 5–6 months.

2.2. Procedures

The children were comfortably seated on a chair with their back supported. They were asked to fix their eyes on a target (a red beak of a Donald Duck drawing) on the wall 90 cm directly in front of their siting position, and to avoid movements of the head. The measures were performed in a silent and comfortable environment. Each recording began with the largest number of teeth in contact. The children were asked to find this starting position by lightly tapping their opposing teeth together and clenching. They were asked to hold this position with a test bolus on the tongue. A number of conditions were then performed which consisted of chewing a soft bolus (chewing gum) and then a hard bolus (wine gum) deliberately on the right and left sides. Each condition lasted for 10 s computer controlled and the children were instructed to chew at a natural pace. Each condition was repeated three times consecutively (total of 3 repetitions for 4 conditions). An operator indicated the side of mastication before each acquisition throughout the session and controlled for its proper execution (visual inspection).

The soft bolus was a piece of chewing gum and the hard bolus was a wine gum, both of which were the same size (20 mm in length, 1.2 mm in height, and 0.5 mm in width) but of different weights (2 g for the soft bolus and 3 g for the hard bolus) and different puncture forces (0.36 N for the soft bolus and 1.85 N for the hard bolus). The wine gum was chosen to provide a rubber-like resistance without sticking to the teeth.

2.3. Kinematic analysis

Mandibular motion was tracked using a Kinesiograph (K7-I; Myotronics, Tukwila, WA, USA) that measures jaw movements with an accuracy of 0.1 mm. Multiple sensors (Hall effect) in a lightweight (113 g) array tracked the motion of a tiny magnet attached at the lower interincisor point (Jankelson, 1980). The Kinesiograph was interfaced with a computer for data storage and subsequent analysis.

2.4. Electromyography (EMG)

Surface EMG signals were recorded from the masseter muscles of both sides using a multichannel EMG amplifier modified with a bandwidth of 45–430 Hz per channel. The EMG amplifier is part of the K7-I WIN Diagnostic System. The relatively large high-pass frequency in EMG recordings was selected to reduce low-frequency movement artifacts during chewing. Two electrodes (Duotrode silver/silver chloride EMG electrodes; Myotronics) were positioned over the masseter muscles bilaterally with an interelectrode distance of 20 mm. Before electrode placement, the skin was cleaned with light abrasive paste and ethanol and the electrodes were positioned along the mandibular angle—cantus straight line to ensure consistency of electrode placement between

sessions (Castroflorio et al., 2005). Kinematic and EMG data were recorded concurrently.

2.5. Signal analysis

The kinematic signals were analyzed using custom-made software (University of Turin, Italy) that allows for automatic data segmentation and analysis. This approach has been described elsewhere (Piancino et al., 2009). The first cycle, during which the bolus was transferred from the tongue to the dental arches, was excluded from the analysis. Jaw movements between two consecutive masticatory pauses were also excluded if they did not represent a chewing cycle based on the presence of at least one of the following characteristics: (i) minimum opening smaller than 4 mm; (ii) duration shorter than 300 ms; or (iii) vertical opening smaller than 3 mm.

From each cycle, the following variables were extracted: (i) cycle duration; (ii) opening duration; (iii) closing duration; (iv) maximum closing velocity; (iv) maximum opening velocity and (v) closure angle. The values computed for each variable were averaged over all cycles recorded for the same side of mastication and the same bolus.

The chewing cycles were divided into non-reverse and reverse, based on the vectorial direction of closure. The closure angle was measured between a straight line obtained by a robust regression procedure on the last part of the curve (from 2.0 to 0.1 mm from the closing point in vertical direction) and the horizontal line of the side of mastication. Next, cycles with a closure angle larger than $90°$ were grouped in the reverse set.

The surface EMG was rectified and low-pass filtered with a 10 Hz cut-off frequency (signal envelope). During each cycle, the maximum values of the EMG envelope of both sides were computed. The percent difference between ipsilateral (deliberate chewing side) and contralateral masseter peak EMG was computed. The percent difference between ipsilateral and contralateral masseter peak EMG was calculated as an indication of the coordination between the bilateral masseter muscles. Such normalization overcomes the known limitations in the use of the EMG amplitude and allows pooling data from different subjects and computing ensemble averages (Piancino et al., 2008).

Muscle onset periods were computed by a wavelet-based method for muscle on–off detection (Merlo, Farina, & Merletti, 2003), which provides accuracy suitable for clinical applications and is completely automatic without any intervention required by the operator. Next, the occlusal pause was calculated as the time difference between the end of the EMG activity of the masseter and the beginning of the next opening phase.

2.6. Statistical analysis

Before comparisons, all variables were tested for normality using the Shapiro-Wilk test and the normal distribution of the data were confirmed. Kinematic and EMG data were analyzed as described previously (Piancino et al., 2009). Firstly, data from the control group were analyzed using repeated-measures analysis of variance (ANOVA) to assess for a potential influence of the side of mastication. No side difference was observed for the control group for the kinematic data thus the data were averaged for mastication on the left and right side. For the children with crossbite, the baseline data from both the reverse and non-reverse cycles were averaged and compared with the data for the non-reverse cycles after the intervention.

To evaluate whether the percentage of reverse cycles changed after the intervention, a three-way ANOVA was applied with time (pre and post intervention), side (crossbite, non-affected), and bolus hardness (soft and hard) as factors. Kinematic variables were

Table 1
Mean values (SD) of kinematic variables during chewing on both the crossbite and the non-crossbite side in the sample patients and in controls. See text for details on data analysis.

	Crossbite affected side				Crossbite non affected side				Controls			
	Pre		Post		Pre		Post		Session 1		Session 2	
	Soft	Hard	Soft	Hard	Soft	Hard	Soft	Hard	Soft	Hard	Soft	Hard
Cycle Duration (ms)	583 (101)	612 (117)	589 (90)	597 (93)	597 (139)	613 (110)	599 (94)	602 (104)	595 (93)	591 (86)	577 (88)	588 (96)
Opening Duration (ms)	228 (57)	236 (57)	240 (55)	238 (57)	241 (69)	249 (64)	252 (58)	246 (63)	224 (25)	224 (37)	235 (46)	253 (54)
Closing Duration (ms)	352 (65)	372 (75)	346 (46)	359 (52)	356 (89)	363 (67)	345 (48)	354 (57)	363 (67)	366 (56)	334 (45)	332 (50)
Maximum Closing Velocity (mm s^{-1})	95 (30)	111 (29)	98 (21)	117 (25)	94 (31)	114 (31)	101 (22)	120 (26)	86 (24)	109 (33)	107 (38)	125 (42)
Maximum Opening Velocity (mm s^{-1})	100 (36)	117 (42)	104 (28)	129 (34)	97 (37)	116 (39)	104 (29)	128 (35)	91 (26)	113 (39)	99 (37)	120 (40)
Closure Angle (deg)	95 (24)	99 (28)	65 (14)	70 (11)	61 (20)	59 (22)	65 (17)	66 (15)	52 (12)	47 (17)	52 (12)	47 (17)

also analyzed with a three-way ANOVA. Factors included in the analysis were group (crossbite, controls), time (pre and post intervention), and bolus hardness (soft and hard). Furthermore, the percent difference between the ipsilateral and contralateral masseter peak EMG was analyzed with a three-way ANOVA by considering group (crossbite, controls), time (pre and post intervention), and bolus hardness (soft and hard) as factors. In addition, the percent change in ipsilateral and contralateral masseter muscle activity pre to post intervention was evaluated in the children with crossbite using a two-way ANOVA with side (ipsilateral, contralateral) and bolus type (soft and hard) as factors.

To identify differences in the mean occlusal pause during mastication on both the affected and non-affected side, a four-way ANOVA was applied with group (crossbite affected side, crossbite non-affected side, controls), time (pre and post intervention), muscle side (ipsilateral and contralateral to the side of mastication/crossbite), and bolus hardness (soft and hard) as factors.

When ANOVA was significant, pair-wise comparisons were tested with the post-hoc Student-Newman-Keuls (SNK) test. The significance level was set at $P < 0.05$.

3. Results

The control subjects yielded $4 \pm 2\%$ (soft bolus) and $5 \pm 3\%$ (hard bolus) reverse cycles at the first evaluation with no significant variations after six months. In the patients with crossbite, the percentage of reverse cycles when chewing on the crossbite side was $57 \pm 30\%$ (soft bolus) and $65 \pm 34\%$ (hard bolus) before the intervention, being significantly greater than in controls ($P < 0.001$), and $12 \pm 17\%$ (soft bolus) and $12 \pm 13\%$ (hard bolus) post intervention. When chewing on the non-affected side, the number of reverse cycles was $15 \pm 19\%$ (soft bolus) and $18 \pm 25\%$ (hard bolus) before the intervention and $12 \pm 18\%$ (soft bolus) and $13 \pm 19\%$ (hard bolus) post intervention. The percentage of reverse cycles were significantly higher when chewing on the crossbite side compared to the non-affected side ($P < 0.001$) and were significantly higher when chewing the hard bolus compared to the soft bolus ($P < 0.05$). The percentage of reverse cycles was significantly reduced after the intervention for both sides and bolus type (main effect of time: $P < 0.001$).

3.1. Kinematics

Table 1 presents the values of the kinematic variables assessed in this study.

The cycle duration was not dependent on group, bolus type or time. Furthermore, the opening and closing duration were not dependent on group, bolus type or time. The maximum velocity of closing was dependent on the bolus type ($P < 0.001$) with faster velocities observed during mastication of a hard bolus however the

maximum velocity of closing was not different between groups and was not affected by the intervention. The maximum velocity of opening was also dependent on the bolus type ($P < 0.001$) with faster velocities observed during mastication of the hard bolus and, similar to the results for closing velocity, the maximum velocity of opening did not differ between groups and was not affected by the intervention.

In contrast, the closing angle was significantly lower after the intervention for the children with crossbite when chewing on the crossbite side for both soft and hard boluses ($P < 0.001$ and $P < 0.005$ respectively). Thus a difference between sides was observed for the children with crossbite at baseline ($P < 0.001$), whereas there was no side difference after intervention ($P = 0.308$; $P = 0.145$ for the soft and hard bolus respectively). No differences were shown during chewing on the non-crossbite side.

3.2. Electromyography

The percent difference between ipsilateral and contralateral masseter peak EMG was dependent on the bolus type ($P < 0.01$) and on the interaction between group and time ($P < 0.05$; Fig. 2). The percent difference between the ipsilateral and contralateral masseter peak EMG was greater for the soft bolus compared to the hard bolus ($P < 0.001$). At baseline the percent difference between the ipsilateral and contralateral masseter peak EMG was lower for
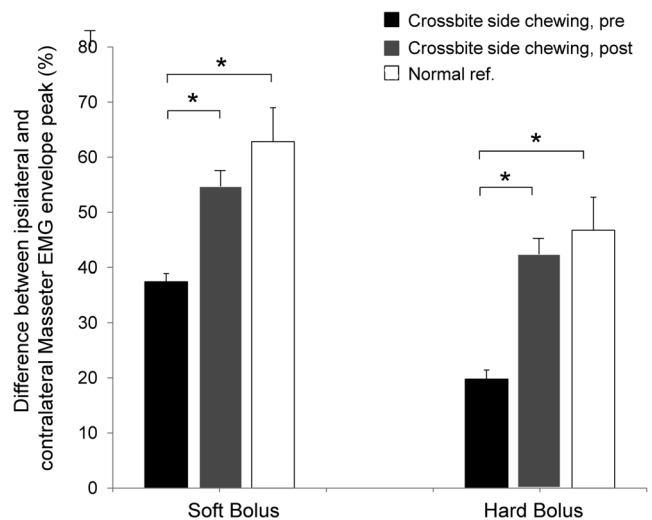

Fig. 2. Mean and SE of the comparison of the percent difference of the ipsilateral and contralateral masseter EMG amplitude during chewing on the crossbite side, soft and hard boluses, before and after therapy, in the patients and control group. * indicates statistically significance difference.

the patients with crossbite compared to the controls however after the intervention there was no difference between groups.

The percent change in ipsilateral and contralateral masseter muscle activity pre to post intervention for both bolus types is presented in Fig. 3. As can be seen in this figure, the normalization of coordination between the bilateral masseter after therapy, was due to a significant reduction of the activity of the contralateral masseter muscle for both bolus types (main effect for side: $P < 0.05$).

The mean occlusal pause was significantly longer for the contralateral masseter compared to the ipsilateral masseter for both groups and bolus types ($P < 0.05$). Furthermore, the mean occlusal pause was significantly longer for the soft versus hard bolus ($P < 0.001$).

An interaction between group and time was observed ($P < 0.01$). That is, independent of the bolus type or side of the masseter muscle, at baseline the mean occlusal pause was lower for the patients with crossbite for mastication on both the affected side ($P < 0.001$) and non-affected side ($P < 0.01$) compared to the control subjects. The mean occlusal pause increased significantly following the intervention for the crossbite patients for both the ipsilateral and contralateral masseter and for both bolus types, however this observation was only present during mastication on the affected side ($P < 0.001$). A trend was present to suggest that
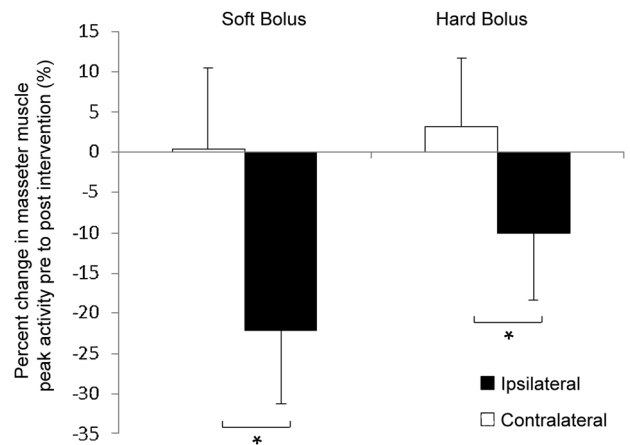
Fig. 3. Percent change in both ipsilateral and contralateral masseter muscle activity pre to post intervention during chewing the standardized soft and hard boli. Ensemble average and standard deviations are presented. The therapy determined a mild percent increase in the ipsilateral EMG activity and a large decrease in the contralateral EMG activity. * indicates statistically significance difference.
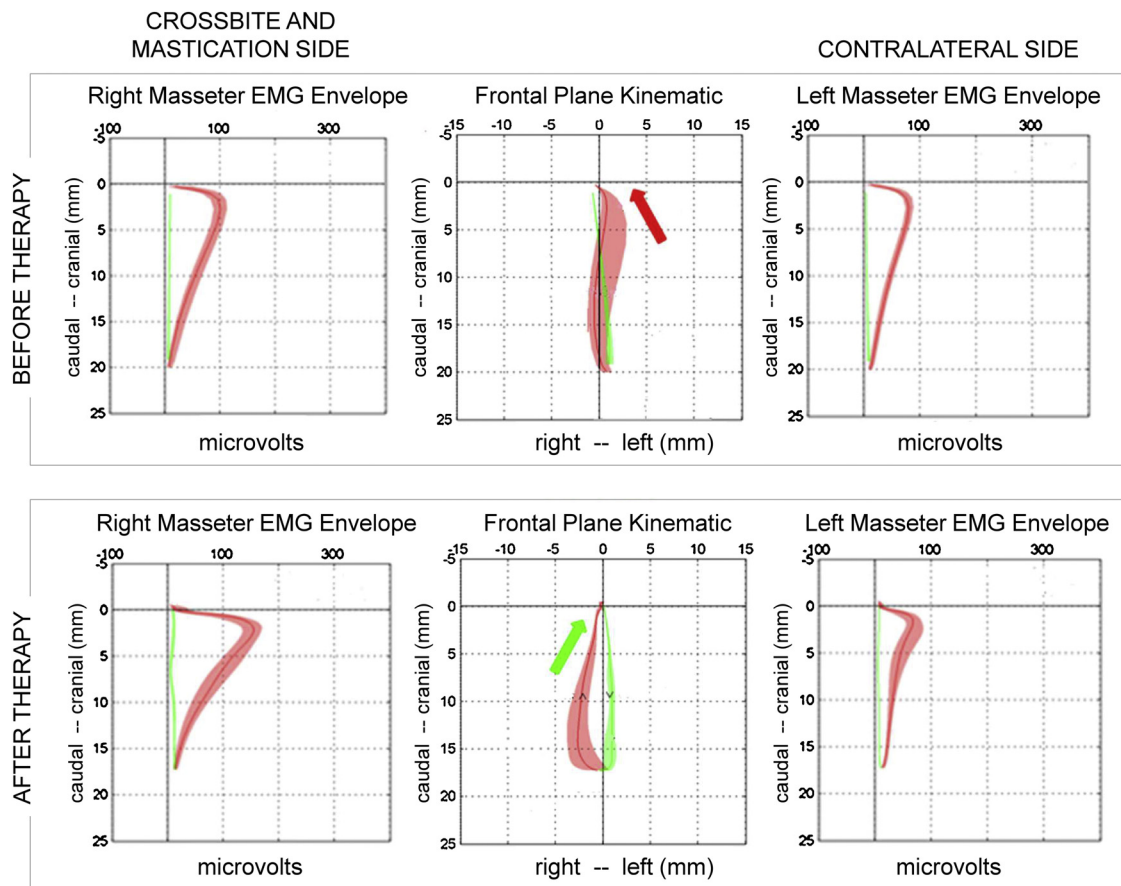
Fig. 4. Masticatory kinematic pattern in the frontal plane (central plot) and EMG envelope plotted versus the vertical jaw displacement of a patient with right posterior unilateral crossbite during chewing of a hard bolus deliberately on the right (crossbite) side before and after functional correction. The solid line, green for the opening and red for the closing pattern, represents the average chewing cycle of 3 trials lasting 10 s each; the green and red areas represent the standard deviation over the average cycle. Top: reverse chewing pattern with a reverse direction of closure (red arrow): note the similarity in the peak EMG envelopes between the two sides, which is not normal during a deliberate side chewing. Below: situation after correction of the malocclusion. Note the normalization of the kinematic chewing pattern in terms of both physiological shape and closing direction (green arrow) along with the recovery of a physiological muscle coordination, which is characterized by a 2:1 ratio between the ipsilateral and the contralateral masseter envelope peaks. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

the mean occlusal pause also increased for mastication on the non-affected side (P = 0.06).

4. Discussion

This study investigated for the first time the effects of functional therapy on masticatory function in patients with unilateral posterior crossbite.

In this work we adopted Lewin's definition of reverse sequence chewing cycles (Lewin & Ramadori, 1985), which refers to the direction of closure, since this has been shown to define the majority of patterns during chewing on the crossbite side (Piancino et al., 2006; Sever et al., 2011; Throckmorton et al., 2001). Furthermore, the reverse sequence chewing cycles have been shown in a decreased activity of the masseter of the crossbite side and in an increased masseter activation of the contralateral side, that is the worse lack of muscular coordination between sides, among the chewing patterns from the crossbite side. As a consequence the masseter muscle of the nonaffected side is more loaded than the masseter muscle of the crossbite side (Piancino et al., 2009).

The activation and coordination of the masseter muscles were evaluated during chewing either soft or hard boluses, on both the affected and non-affected side. Kinematic parameters were also computed to allow comparison of kinematic patterns obtained from our sample with those already available in the literature. Before therapy, the percentage of reverse sequencing chewing cycles, according to the Lewin's definition (Lewin & Ramadori, 1985), and the closing angle showed differences between the affected and unaffected side, which is in agreement with previous observations (Ben-Bassat et al., 1993; Piancino et al., 2013; Throckmorton et al., 2001), thus ensuring data quality and validity.

Since the amplitude of the EMG signal is influenced by individual anatomical characteristics, this study compared the percent difference of the ipsilateral and contralateral masseter EMG amplitude as a way of normalizing the data. The percent difference in masseter muscle activity between sides in children with posterior crossbite when chewing on the crossbite side was significantly lower compared to the difference observed for the control group and during chewing on the normal side, corresponding to the altered kinematics of the reverse chewing cycles (Andrade et al., 2010; Piancino et al., 2009).

After the intervention, a significant reduction in reverse chewing cycles was observed when chewing on the previous crossbite side and the angle of closure was no longer different between sides for either bolus type. Furthermore, the percent difference in masseter muscle activity between sides was similar for the patients after therapy to that of control subjects, thus indicating that the intervention had induced a symmetrization and a favorable change in the neuromuscular control of chewing (Fig. 4). This was also observed for both bolus types. The normalization of muscle activity between sides after the intervention corresponds to the symmetry observed for the kinematic data post treatment. In particular, the reduction of the percentage of reverse chewing cycles is an important sign of the restoration of the coordination of bilateral masseter muscle activity (Piancino et al., 2009; Tomonari, Ikemori, Kubota, Uehara, & Miyawaki, 2014; Tomonari, Kubota et al., 2014).

The mean occlusal pause was lower for the patients with crossbite during chewing on both sides (Sever et al., 2011). Interestingly, the mean occlusal pause became significantly longer, after the intervention, when chewing on the affected side. As a result, the mean occlusal pause was comparable between groups for both masseter muscles and during chewing with both bolus types.

The appliance used in this study controls the position of the mandible in 3-dimensional space preventing the upper and lower teeth from cusp to cusp contact during the orthodontic movements. This might be an important aspect for restoration of masticatory function.

Collectively, these findings show that masticatory function is seriously affected in patients with unilateral posterior crossbite and that, post intervention with Function Generating Bite appliance, both the kinematics and the muscular activation more closely reflect that of a control group. The achievement of this outcome is important to avoid the potential overloading and imbalance of muscles and structures which may occur when asymmetry is present.

5. Conclusion

The correction of the malocclusion with a functional appliance induced a favorable change in the neuromuscular control of chewing of patients, who recovered a normal-like coordination between the masseter muscles during chewing and a significant reduction of the reverse chewing patterns. The altered muscular activation corresponded to the altered kinematics of reverse chewing cycles that might be considered a useful indicator of the severity of the masticatory function involvement.

Competing interest

The authors declare that they have no conflict of interest.

Author contributions

M.G. Piancino contributed to conception, design and data interpretation, drafted and critically revised the manuscript; D. Falla contributed to design, data analysis and interpretation, and critically revised the manuscript; A. Merlo contributed to design, data analysis and interpretation, and critically revised the manuscript; T. Vallelonga contributed to design, data acquisition and interpretation; C. de Biase contributed to design, data acquisition and interpretation, drafted and critically revised the manuscript; D. Dalessandri contributed to design and critically revised the manuscript; C.L. Debernardi contributed to design and critically revised the manuscript. All authors gave final approval of the version to be submitted.

Ethical approval

The protocol in this study was approved by the Institutional Review Board of the University Hospital "Health and Science Complex Turin-Italy" n. CS/246.

References

Ahlgren, J. (1967). Pattern of chewing and malocclusion of teeth. A clinical study. *Acta Odontologica Scandinavica*, *25*(1), 3–13 Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/5233858.

Andrade, S., Gavião, M. B. D., Gameiro, G. H., & De Rossi, M. (2010). Characteristics of masticatory muscles in children with unilateral posterior crossbite. *Brazilian*

Oral Research, 24(2), 204–210 Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/20658040.

Ben-Bassat, Y., Yaffe, A., Brin, I., Freeman, J., & Ehrlich, Y. (1993). Functional and morphological-occlusal aspects in children treated for unilateral posterior cross-bite. *European Journal of Orthodontics*, 15(1), 57–63 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/8436197.

Castroflorio, T., Farina, D., Bottin, A., Piancino, M. G., Bracco, P., & Merletti, R. (2005). Surface EMG of jaw elevator muscles: effect of electrode location and inter-electrode distance. *Journal of Oral Rehabilitation*, 32(6), 411–417. http://dx.doi.org/10.1111/j.1365-2842.2005.01442.x.

Da Silva Filho, O. G., Santamaria, M., & Capelozza Filho, L. (2007). Epidemiology of posterior crossbite in the primary dentition. *The Journal of Clinical Pediatric Dentistry*, 32(1), 73–78 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/18274476.

Ferrario, V. F., Sforza, C., & Serrao, G. (1999). The influence of crossbite on the coordinated electromyographic activity of human masticatory muscles during mastication. *Journal of Oral Rehabilitation*, 26(7), 575–581 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/10445477.

Jankelson, B. (1980). Measurement accuracy of the mandibular kinesiograph—a computerized study. *The Journal of Prosthetic Dentistry*, 44(6), 656–666 Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/6934297.

Johnsen, S. E., & Trulsson, M. (2005). Encoding of amplitude and rate of tooth loads by human periodontal afferents from premolar and molar teeth. *Journal of Neurophysiology*, 93(4), 1889–1897. http://dx.doi.org/10.1152/jn.00664.2004.

Kiliaridis, S., Mahboubi, P. H., Raadsheer, M. C., & Katsaros, C. (2007). Ultrasonographic thickness of the masseter muscle in growing individuals with unilateral crossbite. *The Angle Orthodontist*, 77(4), 607–611. http://dx.doi.org/10.2319/101105-360.

Lewin, A., & Ramadori, G. (1985). *Electrognathographics: atlas of diagnostic procedures and interpretation*. Chicago: Quintessence Pub. Co..

Lund, J. P., Scott, G., Kolta, A., & Westberg, G. R. (1999). Role of cortical inputs and brainstem interneuron populations in patterning mastication. In Y. Nakamura, & G. J. Sessle (Eds.), *Neurobiology of mastication. From molecular to system approach* (pp. 504–514).Tokyo: Elsevier Science.

Martín, C., Palma, J. C., Alamán, J. M., Lopez-Quiñones, J. M., & Alarcón, J. A. (2012). Longitudinal evaluation of sEMG of masticatory muscles and kinematics of mandible changes in children treated for unilateral cross-bite. *Journal of Electromyography and Kinesiology*, 22(4), 620–628. http://dx.doi.org/10.1016/j.jelekin.2012.01.002.

Merlo, A., Farina, D., & Merletti, R. (2003). A fast and reliable technique for muscle activity detection from surface EMG signals. *IEEE Transactions on Bio-Medical Engineering*, 50(3), 316–323. http://dx.doi.org/10.1109/TBME.2003.808829.

Morquette, P., Lavoie, R., Fhima, M.-D., Lamoureux, X., Verdier, D., & Kolta, A. (2012). Generation of the masticatory central pattern and its modulation by sensory feedback. *Progress in Neurobiology*, 96(3), 340–355. http://dx.doi.org/10.1016/j.pneurobio.2012.01.011.

Nerder, P. H., Bakke, M., & Solow, B. (1999). The functional shift of the mandible in unilateral posterior crossbite and the adaptation of the temporomandibular joints: a pilot study. *European Journal of Orthodontics*, 21(2), 155–166 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/10327739.

Piancino, M. G., Talpone, F., Dalmasso, P., Debernardi, C., Lewin, A., & Bracco, P. (2006). Reverse-sequencing chewing patterns before and after treatment of children with a unilateral posterior crossbite. *European Journal of Orthodontics*, 28(5), 480–484. http://dx.doi.org/10.1093/ejo/cjl014.

Piancino, M. G., Bracco, P., Vallelonga, T., Merlo, A., & Farina, D. (2008). Effect of bolus hardness on the chewing pattern and activation of masticatory muscles in subjects with normal dental occlusion. *Journal of Electromyography and Kinesiology*, 18(6), 931–937. http://dx.doi.org/10.1016/j.jelekin.2007.05.006.

Piancino, M. G., Farina, D., Talpone, F., Merlo, A., & Bracco, P. (2009). Muscular activation during reverse and non-reverse chewing cycles in unilateral posterior crossbite. *European Journal of Oral Sciences*, 117(2), 122–128. http://dx.doi.org/10.1111/j.1600-0722.2008.00601.x.

Piancino, M. G., Frongia, G., Dalessandri, D., Bracco, P., & Ramieri, G. (2013). Reverse cycle chewing before and after orthodontic-surgical correction in class III patients. *Oral Surgery, Oral Medicine, Oral Pathology and Oral Radiology*, 115(3), 328–331. http://dx.doi.org/10.1016/j.oooo.2012.04.025.

Pinto, A. S., Buschang, P. H., Throckmorton, G. S., & Chen, P. (2001). Morphological and positional asymmetries of young children with functional unilateral posterior crossbite. *American Journal of Orthodontics and Dentofacial Orthopedics*, 120(5), 513–520. http://dx.doi.org/10.1067/mod.2001.118627a.

Pirttiniemi, P., Kantomaa, T., & Lahtela, P. (1990). Relationship between craniofacial and condyle path asymmetry in unilateral cross-bite patients. *European Journal of Orthodontics*, 12(4), 408–413 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/2086261.

Proffit, W. R., Fields, H. W., & Moray, L. J. (1998). Prevalence of malocclusion and orthodontic treatment need in the United States: estimates from the NHANES III survey. *The International Journal of Adult Orthodontics and Orthognathic Surgery*, 13(2), 97–106 Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/9743642.

Quintero, A., Ichesco, E., Schutt, R., Myers, C., Peltier, S., & Gerstner, G. E. (2013). Functional connectivity of human chewing: an fcMRI study. *Journal of Dental Research*, 92(3), 272–278. http://dx.doi.org/10.1177/0022034512472681.

Rilo, B., Silva, J. L., da Mora, M. J., Cadarso-Suárez, C., & Santana, U. (2007). Unilateral posterior crossbite and mastication. *Archives of Oral Biology*, 52(5), 474–478. http://dx.doi.org/10.1016/j.archoralbio.2006.10.001.

Sever, E., Marion, L., & Ovsenik, M. (2011). Relationship between masticatory cycle morphology and unilateral crossbite in the primary dentition. *European Journal of Orthodontics*, 33(6), 620–627. http://dx.doi.org/10.1093/ejo/cjq070.

Thilander, B., & Bjerklin, K. (2012). Posterior crossbite and temporomandibular disorders (TMDs): need for orthodontic treatment? *European Journal of Orthodontics*, 34(6), 667–673. http://dx.doi.org/10.1093/ejo/cjr095.

Thilander, B., & Lennartsson, B. (2002). A study of children with unilateral posterior crossbite, treated and untreated, in the deciduous dentition. *Journal of Orofacial Orthopedics/Fortschritte Der Kieferorthopädie*, 63(5), 371–383. http://dx.doi.org/10.1007/s00056-002-0210-6.

Throckmorton, G. S., Buschang, P. H., Hayasaki, H., & Pinto, A. S. (2001). Changes in the masticatory cycle following treatment of posterior unilateral crossbite in children. *American Journal of Orthodontics and Dentofacial Orthopedics*, 120(5), 521–529. http://dx.doi.org/10.1067/mod.2001.118626.

Tomonari, H., Ikemori, T., Kubota, T., Uehara, S., & Miyawaki, S. (2014). First molar cross-bite is more closely associated with a reverse chewing cycle than anterior or pre-molar cross-bite during mastication. *Journal of Oral Rehabilitation*, 41(12), 890–896. http://dx.doi.org/10.1111/joor.12222.

Tomonari, H., Kubota, T., Yagi, T., Kuninori, T., Kitashima, F., Uehara, S., et al. (2014). Posterior scissors-bite: masticatory jaw movement and muscle activity. *Journal of Oral Rehabilitation*, 41(4), 257–265. http://dx.doi.org/10.1111/joor.12148.

Wilding, R. J., & Lewin, A. (1994). The determination of optimal human jaw movements based on their association with chewing performance. *Archives of Oral Biology*, 39(4), 333–343 Retrieved from http://www.ncbi.nlm.nih.gov/ubmed/8024498.

Woda, A., Nicolas, E., Mishellany-Dutour, A., Hennequin, M., Mazille, M.-N., Veyrune, J.-L., et al. (2010). The masticatory normative indicator. *Journal of Dental Research*, 89(3), 281–285. http://dx.doi.org/10.1177/0022034509357022.