



Diaphragmatic amplitude and accessory inspiratory muscle activity in nasal and mouth-breathing adults: A cross-sectional study



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ABSTRACT

The purpose of this study was to evaluate the electromyographic activity of the accessory inspiratory muscles and the diaphragmatic amplitude (DA) in nasal and mouth-breathing adults. The study evaluated 38 mouth-breathing (MB group) and 38 nasal-breathing (NB group) adults, from 18 to 30 years old and both sexes. Surface electromyography (sEMG) was used to evaluate the amplitude and symmetry (POC%) of the sternocleidomastoid (SCM) and upper trapezius (UT) muscles at rest, during nasal slow inspiration at Lung Total Capacity (LTC) and, during rapid and abrupt inspiration: Sniff, Peak Nasal Inspiratory Flow (PNIF) and Maximum Inspiratory Pressure (MIP). M-mode ultrasonography assessed the right diaphragm muscle amplitude in three different nasal inspirations: at tidal volume (TV), Sniff and inspiration at LTC. The SCM activity was significantly lower in the MB group during Sniff, PNIF ($p < 0.01$, Mann–Whitney test) and MIP ($p < 0.01$, t -test). The groups did not differ during rest and inspiration at LTC, regarding sEMG amplitude and POC%. DA was significantly lower in the MB group at TV ($p < 0.01$, Mann–Whitney) and TLC ($p = 0.03$, t -test). Mouth breathing reflected on lower recruitment of the accessory inspiratory muscles during fast inspiration and lower diaphragmatic amplitude, compared to nasal breathing.

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1. Introduction

Nasal breathing is innate to human beings due to its important function of preparing the air to reach the important structures of the respiratory system. In addition, it is essential for development and functionality of the craniofacial and stomatognathic systems (Armijo-Olivo et al., 2006; Cuccia et al., 2008).

Nasal breathing can be partially or totally replaced by mouth breathing, regardless of the factors that block the passage of the air through the nasopharynx. These factors can be obstructive, mainly the enlarged adenotonsillar tissues, or functional, caused by transient edema of nasal mucosa, muscular flaccidity or by the maintenance of this habit even after surgical correction (Berwig et al., 2011). Mouth supplying is considered an abnormal and inefficient adaptation of breathing mode and it may induce

functional, postural, biomechanical and occlusal imbalances (Barros et al., 2006; Okuro et al., 2011).

Mouth-breathing (MB) mode, as obstructive as functional, may produce postural adaptations and muscular imbalances in the attempt of reducing nasal resistance and facilitating airflow through the nasal-pharyngeal airway. Forward head posture is commonly found in MB people (Chaves et al., 2010; Cuccia et al., 2008; Okuro et al., 2011; Yi et al., 2008), reflecting on the diaphragm (Lima et al., 2004; Yi et al., 2008) and rib cage kinetics (Okuro et al., 2011; Pires et al., 2007). Additionally, studies found higher accessory inspiratory muscle activity at rest (Ribeiro et al., 2002; Corrêa and Bérzin, 2008), respiratory muscle weakening (Milanesi et al., 2014; Okuro et al., 2011; Pires et al., 2005, 2007) and predominant inspiratory movement in the upper thorax (Yi et al., 2008). The lower functional exercise capacity and the reduced quality of life in the domain of general health were also consequences of the mouth-breathing mode (Milanesi et al., 2014).

Sternocleidomastoideus and upper trapezius muscles are described as inspiratory accessory muscles, acting as head and

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neck stabilizers and helping to elevate the thoracic cage, respectively, during deep inspiration (Kendall, 2005). Studies conducted with sEMG found higher activity of the accessory inspiratory muscles at rest, and lower activity during maximal contraction, in MB children compared to the nasal breathers. It was suggested that this activity pattern may occur due to muscular imbalance, resulting from postural changes in mouth-breathing children (Ribeiro et al., 2002). In another study carried out with MB children (Corrêa and Bérzin, 2008), the EMG activity levels of the accessory inspiratory muscles, previously to physiotherapeutic intervention, were compatible with muscular hyperactivity.

Diaphragmatic amplitude (DA) in MB children, investigated by means of fluoroscopy (Yi et al., 2008), was lower than in nasal-breathing children. Traditionally diaphragm evaluation is accomplished by fluoroscopy (gold pattern); however, ultrasound has shown advantages due to absence of radiation and portability (Boussuges et al., 2009; Nason et al., 2012). Despite the different approaches, either directly attained at the posterior surface of the diaphragm (Kim et al., 2010) or indirectly (craniocaudal displacement of the left branch of the portal vein) (Grams et al., 2014; Yamaguti et al., 2007), there is a consensus that ultrasound is a reproducible and easy to use method (Boussuges et al., 2009; Grams et al., 2014; Kim et al., 2010; Nason et al., 2012).

Currently, most studies regarding mouth-breathing mode address otorhinolaryngological, dental and orofacial motricity aspects, however, some of them concentrate on the postural changes, mainly in children. Nevertheless, morphofunctional sequelae of these changes may remain at adult age, even after treating the nasal obstruction during youth.

The evaluation and treatment of mouth breathing, by a physical therapist, should comprehend postural and ventilatory changes. However, there is scarcity of studies regarding ventilatory function and respiratory muscles in mouth-breathing children and adults.

The knowledge obtained concerning diaphragmatic motion and accessory inspiratory muscles recruitment, in mouth-breathing adults, may contribute to a more global and interdisciplinary diagnostic and therapeutic approach, from childhood to adult age.

The objective of this study was to evaluate the electrical activity of accessory inspiratory muscles and the amplitude of diaphragmatic movement in mouth and nasal-breathing adults.

2. Methods

This exploratory, cross-sectional, controlled study was approved by the local Research Ethics Committee (protocol number 04039912.7.0000.5346) and a Consent Form was signed by the participants. Adult male and females between 18 and 30 years of age with mouth-breathing (MB group) and nasal-breathing mode (NB group), body mass index (BMI) between 18.5 and 24.9 kg/m² (WHO, 2013), normal spirometry according to European Respiratory Society (Stocks and Quanjer, 1995) and without evidence of respiratory and neuromuscular diseases, thoracic deformities, tobacco history and/or exposure to risk environment, took part in the study. Subjects using topical or systemic corticosteroids, muscle relaxants and/or barbiturates were excluded. Those with report of flu in the last three weeks or allergic rhinitis attack on the assessment day, chest and/or abdominal surgery, abdominal hernia and those who were physically active were excluded. The diagnosis of the breathing mode was based on the anamnesis, signs and symptoms and physical characteristics related to mouth breathing, such as absence of lip seal, hypotonic lips, elongated face, report of snoring, drooling on the pillow, or having the mouth open most part of the day and/or during sleep (Milanesi et al., 2014; Yi et al., 2008). The otorhinolaryngologic evaluation included nasofibroscope, which assessed the presence of upper airway

obstruction by tonsillar and or adenoidal hypertrophy or rhinitis and classified the participants as organic or functional mouth breathers, namely with or without mechanical obstruction.

The anamnesis consisted of collecting signs and symptoms, history of surgery, consumption of drugs, demographic and anthropometric data, stature, body mass index (Digital Scale CAMRY, model EB9013), physical activity level (International Physical Activity Questionnaire – IPAQ) (Matsudo et al., 2001) and spirometry (One Flow FVC KIT, Clement Clark International, United Kingdom). Subsequently, participants were evaluated by sEMG and diaphragm ultrasound.

During examination the subjects were seated with an upright trunk, relaxed arms, hands on lap, eyes open, head in the Frankfurt plane, and the feet flat on the floor. For the sEMG, an adapted chair was used (Corrêa and Bérzin, 2008).

Electrical activity of Sternocleidomastoideus (SCM) and upper trapezius (UT) muscles was recorded bilaterally. Signal acquisition was based on the *Surface ElectroMyoGraphy for the Non-Invasive Assessment of Muscles* (SENIAM) (Hermens et al., 2000). In order to reduce signal interference, the collection site was equipped with a rubber floor, and all bulbs and mobile phones were switched off. Any metal accessories were also removed from the participants.

Prior to attaching the electrodes the participants skin was cleaned with gauze soaked with 70% alcohol and, whenever necessary, trichotomy was carried out on the muscle surface.

Preamplifier sensors with differential output were connected to adhesive disposable silver/silver chloride bipolar surface electrodes (interelectrode distance 20 mm; Hall Industry and Commerce Ltda). A reference electrode (Meditrace 100) was applied to the sternal manubrium (Hermens et al., 2000).

The SCM electrodes were positioned longitudinally to the muscle fibers at the midpoint of the muscular belly, located by muscle palpation during manually resisted neck flexion. The UT electrodes were positioned at the midpoint between the spinous process of the seventh cervical vertebra and the acromion (Hermens et al., 2000).

An sEMG signal was acquired using a 14-bit Surface Electromyograph (Miotool 400, Miotec[®], Porto Alegre, Brazil), 110 dB Common Mode Rejection and 2000 Hz sampling frequency per channel. Butterworth and band-pass filters (20–500 Hz) were used (Hermens et al., 2000). A signal was recorded by Miograph (Miotec[®], Porto Alegre, Brazil) and stored in a portable computer (HP 420 Intel Celeron). Both devices were connected to battery without connection to a power system.

Muscle activity was recorded at rest (tidal volume – TV), during 10 s, and during four inspiratory tests: Sniff, Lung Total Capacity (LTC), Peak Nasal Inspiratory Flow (PNIF) and Maximum Inspiratory Pressure (MIP). These tests started after expiration at Functional Residual Capacity (FRC), except the MIP, which started from Residual Volume (RV). From these expiratory levels, the subjects performed fast and abrupt nasal inspiration (Sniff), slow and complete nasal inspiration (LTC), fast nasal inspiration through a facial mask (PNIF) and fast, forced and sustained inspiration through the mouthpiece (MIP) (Kjærgaard et al., 2008; Montemezzo et al., 2012; Neder et al., 1999; Ottaviano et al., 2008). Each of these activities was performed at least three times with a 2-min interval. To normalize the EMG signal, muscle activity during Maximal Voluntary Contraction (MVC) was collected during anterior head flexion (SCM muscle) and shoulder elevation (UT), and with resistance offered by existing boards in the adapted chair (Corrêa and Bérzin, 2008). These activities were recorded during five seconds and repeated at least twice, with 2-min intervals (Figs. 1 and 2).

From all recordings, the most uniform signal that was visually verified and confirmed by the Fast Fourier Transform (FFT) curve was selected for further quantitative analysis (Corrêa and Bérzin,

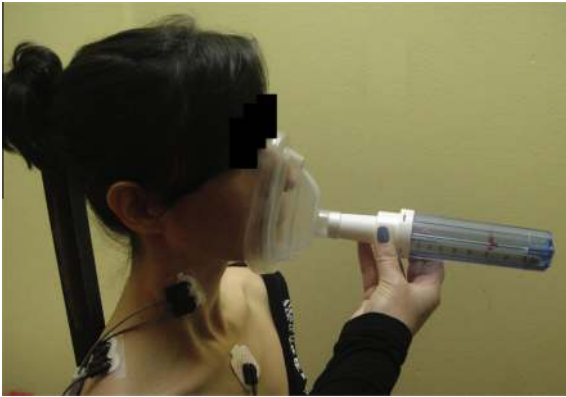


Fig. 1. EMG during measurement of Peak Nasal Inspiratory Flow.

2008). For the Sniff test, 500 ms of the signal was cut, while for others, one second with the highest EMG amplitude was cut. Muscle activity was measured on an amplitude domain, represented by the Root Mean Square (RMS), expressed in microvolt (μV) and transformed into relative values from a reference defined as 100% (MVC).

The Percentage of Overlapping Coefficient (POC%), obtained by overlapping of the normalized values of the amplitude EMG signal from the right and left side, indicates how symmetric the muscle action is. When both sides contract in perfect symmetry and amplitude curves completely overlap, the POC value is 100% (Bouffleur et al., 2014; Ferrario et al., 2000; Ries et al., 2008).

The EMG potentials were filtered by a band-pass filter with 20–50 Hz bandwidth and POC% was obtained by RMS mean potential using a moving window of 25 ms (50 data). EMG data were processed by MATLAB software (*The MathWorks*® version 7.6.0 R2008a).

Diaphragmatic amplitude was verified by ultrasound (US), performed in a platform HD11 (Philips Medical System; Bothel, WA, EUA) with a 3–6 Hz convex transducer. Subjects were assessed by the end of the morning, 2–3 h after a light meal.

The transducer was positioned below the right costal margin, between the midclavicular and anterior axillary lines, using the liver as an acoustic window (Boussuges et al., 2009; Gerscovich et al., 2001; Gierada et al., 1998; Scott et al., 2006). Bidimensional mode (B-mode) was utilized to select the echogenic line of the right hemidiaphragm (Boussuges et al., 2009) and M-mode for diaphragm movement recording (Kim et al., 2010).



Fig. 2. EMG during Maximal Voluntary Contraction of the SCM muscle in an adapted chair.

Three inspiratory tests, previously demonstrated and practiced, were performed in the following sequence: nasal inspiration at TV, Sniff and inspiration at LTC. Tests started and finished at FRC and were repeated at least three times or until three technically satisfactory measures had been obtained, within a 30-s interval (Kim et al., 2010).

With the first ultrasonic caliper placed on the base of the inspiratory curve of the echoic line and the second caliper in the curve apex, diaphragmatic movement amplitude was calculated, i.e., the distance (in cm) between these points (Kim et al., 2010). The EMG and the US assessments were carried out at different moments.

The study sample size was calculated considering a 5% ($p < 0.05$) significance level (alpha) and 80% power (1-beta) (*software WinPepi*, version 1.5). By the greatest variability obtained in a pilot study with 10 subjects, data from diaphragm ultrasound during TV was considered. Thus, 19 subjects at least were to be analyzed in each group.

For statistics analysis, *SPSS software* (17.0) was used, with 5% of significance level and Shapiro–Wilk as a normality test. The groups comparison was analyzed by the *t*-student test for data with a normal distribution and the Mann Whitney test for non-normal distribution. The effect size (*d*) was calculated by a model proposed by Cohen, for mean difference of two independent groups, with *d* value < 0.2 representing a small effect, a medium effect from 0.2 to 0.8 and a large effect $d \geq 0.8$ (Cohen, 1992).

3. Results

The study was conducted with 77 volunteers, 39 (28 women and 11 men) in the NB group and 38 (25 women and 13 men) in the MB group, with a mean age of 22.6 ± 2.9 and 22.7 ± 3.5 years old, respectively. There was no difference between groups related to age ($p = 0.99$), body mass ($p = 0.05$), stature ($p = 0.45$), and BMI ($p = 0.05$). Both groups presented normal spirometric values and were physically non-active. According to the otorhinolaryngologic examination, all participants of the MB group were classified as functional, that is, they breathe through the mouth, although the upper airways did not present mechanical obstruction.

The EMG activity of the accessory inspiratory muscles did not differ between MB and NB groups at rest and LTC, except the left UT muscle which presented significantly higher activity at LTC in the MB group. Nevertheless, this result had low statistical power (34%). In other tests (Sniff, PNIF and MIP), the SCM activity was lower in the MB group (Table 1).

For the muscle symmetry index (POC%), there was no difference between groups in all tests, except the UT muscle which showed significantly lower symmetry at rest in the NB group, but with low statistical power (53%) (Table 2). The mean value of the MIP in the MB group was significantly lower than in the NB group ($91 \pm 14.84\%$ and $100.18 \pm 14.51\%$, respectively).

In DA assessment, smaller amplitude in the MB group was observed in all tests, with significant differences at TV and LTC (Table 3).

The statistical power was high (approximately 80%) and the effect size was medium (0.63–0.74) in the EMG results of the SCM muscle during Sniff, PNIF and MIP and in the DA at TV, supporting the statistical difference between groups.

4. Discussion

The present study, conducted with adults, evaluated the effects of mouth-breathing mode under a relatively novel research perspective, making the comparison with the literature findings difficult. Findings concerned with diaphragmatic amplitude and

Table 1
EMG normalized values (%) of the accessory inspiratory muscles in MB and NB groups.

Activity	Muscles	NB group (n = 39) Mean ± SD	MB group (n = 38) Mean ± SD	p-value
Rest	R SCM	2.09 ± 1.14	1.83 ± 1.00	0.36
	L SCM	1.99 ± 1.10	2.04 ± 1.73	0.55
	R UT	2.33 ± 2.08	2.75 ± 2.40	0.32
	L UT	2.38 ± 1.91	3.68 ± 3.24	0.07
LTC	R SCM	14.04 ± 10.51	16.70 ± 15.91	0.63
	L SCM	13.81 ± 12.46	16.29 ± 15.05	0.38
	R UT	3.90 ± 2.69	5.66 ± 5.25	0.18
	L UT ^b	3.95 ± 3.68	5.38 ± 4.33	<0.01*
Sniff	R SCM ^b	66.22 ± 38.84	40.83 ± 29.51	<0.01*
	L SCM ^b	70.66 ± 46.44	41.78 ± 36.43	<0.01*
	R UT	10.46 ± 12.29	7.37 ± 5.48	0.56
	L UT	9.48 ± 9.69	7.39 ± 4.97	0.65
PNIF	R SCM ^b	109.19 ± 57.78	75.23 ± 46.26	<0.01*
	L SCM ^b	101.16 ± 46.05	71.43 ± 43.59	<0.01*
	R UT	13.32 ± 12.44	13.37 ± 12.33	0.94
	L UT	11.28 ± 9.35	12.02 ± 10.33	0.71
MIP	R SCM ^a	98.18 ± 46.79	70.98 ± 37.88	<0.01*
	L SCM ^a	93.05 ± 49.23	65.88 ± 32.29	<0.01*
	R UT	17.19 ± 16.74	21.10 ± 18.30	0.80
	L UT	15.18 ± 13.16	16.30 ± 15.19	0.85

SD: standard deviation; LTC: Lung Total Capacity; Sniff: fast and short inspiration; PNIF: Peak Nasal Inspiratory Flow; MIP: maximal inspiratory pressure; SCM: Sternocleidomastoideus; UT: upper trapezius; R: right; L: left.

* Significant difference.

^a T-test for independent data.

^b Mann–Whitney test.

electrical activity of accessory inspiratory muscles were compared to data obtained in studies with MB children (Corrêa and Bérzin, 2008; Lima et al., 2004; Pires et al., 2005; Yi et al., 2008), MB and healthy adults (Milanesi et al., 2011, 2014; Boussuges et al., 2009; Costa et al., 1997; Gierada et al., 1998; Hruska, 1997; Testa et al., 2011; Tomich et al., 2007; Yamaguti et al., 2007) or other studies involving EMG of neck muscles (Ries et al., 2008; Sforza et al., 2011; Tartaglia et al., 2008).

EMG of the accessory inspiratory muscles showed that signal amplitude increases with increasing speed and load increment, as occurred during fast inspiration tests (Sniff, PNIF and MIP). Some authors (Costa et al., 1997; Tomich et al., 2007) have attributed the highest activity to the load increment imposed on the respiratory muscles because of the increasing speed of inspiratory flow. In order to overcome this load, they needed to activate a greater number of motor units (Harridge, 2007; He et al., 2000).

Table 2
Muscle symmetry index (POC%) between pairs of evaluated muscles in NB and MB groups.

Variables		NB group (n = 39) Mean ± SD	MB group (n = 38) Mean ± SD	p-value
Rest	SCM	84.03 ± 12.48	84.43 ± 12.15	0.94
	UT ^a	71.52 ± 21.61	80.26 ± 14.98	0.04*
LTC	SCM	81.85 ± 15.61	83.26 ± 13.27	0.79
	UT	80.26 ± 18.57	80.01 ± 16.89	0.78
Sniff	SCM	86.81 ± 12.21	83.58 ± 11.65	0.11
	UT	79.06 ± 17.55	81.70 ± 12.52	0.81
PNIF	SCM	87.93 ± 8.90	83.63 ± 13.53	0.24
	UT	80.35 ± 16.87	82.67 ± 12.77	0.57
MIP	SCM	84.11 ± 15.39	87.44 ± 10.94	0.35
	UT	75.49 ± 22.44	84.87 ± 11.06	0.17

SD: standard deviation; LTC: Lung Total Capacity; Sniff: fast and short inspiration; PNIF: Peak Nasal Inspiratory Flow; MIP: maximal inspiratory pressure; SCM: Sternocleidomastoideus; UT: upper trapezius; R: right; L: left.

* Significant difference.

^a T-test for independent data.

In addition to this, exercises of short duration and high intensity involve mostly type II fibers, characterized by fast contraction and less fatigue resistance, predominant in the accessory inspiratory muscles (He et al., 2000; Polla et al., 2004).

Similarly to previous studies (Costa et al., 1997; Tomich et al., 2007), this study evidenced greater activation of SCM muscle during fast and short inspiratory efforts, which involve higher work load, in both groups, however, it was lower in MB group. The EMG activity level observed in the NB group was similar to the one found in healthy adults (Costa et al., 1997), while MB subjects presented lower muscle recruitment during fast inspiration. This finding may be explained by the fact that the MB subjects, in addition to a greater work load during fast inspiration, underwent an overload imposed by a possible transient edema of nasal mucosa.

Another explanation for the results is the influence of breathing mode to the craniocervical posture. In forward head posture, commonly observed in MB subjects (Milanesi et al., 2011; Yi et al., 2008), cervical muscles may be in a mechanical disadvantage to produce force, due to changes on muscle length-tension relation (Correia, 2012). According to this relation, there is an optimal length for the force production, next to the resting length. Lower or higher than this length, i.e., when the sarcomere is excessively stretched or shortened, force production reduces as a result of smaller actin and myosin myofilaments overlapping and a reduced number of cross-bridges (Correia, 2012). Besides postural impairment, mouth breathing requires less muscular effort, produces inhibition of nasal afferent impulses (van Spronsen et al., 2008) and affects respiratory muscle action with progressive force reduction (Milanesi et al., 2014; Pires et al., 2005).

The lower inspiratory pressure (MIP) observed in MB subjects reinforces the hypothesis that the accessory inspiratory muscles are at a disadvantage in muscle length-tension relation, resulting in lower EMG activation.

The lack of reference values for the muscular symmetry index (POC%) makes the interpretation of the results hard. Similar POC% values were observed in the SCM and UT muscles in control subjects (Ries et al., 2008; Tartaglia et al., 2008; Sforza et al., 2011) compared to the values observed during rest, Sniff, PNIF and MIP, in both groups evaluated in the present study. Such results showing symmetric activity in neck muscles were unexpected, once the possible postural changes, mainly in the head and neck related to MB, could yield muscular unbalance and, consequently, less muscular symmetry in this group.

Diaphragmatic amplitude was significantly lower during inspiration in TV and LTC in the MB group. Nevertheless, compared to results obtained in 200 healthy subjects (Boussuges et al., 2009) in the orthostatic position, they were higher at rest and lightly reduced at LTC in both groups.

During LTC, higher values of DA (7–11 cm) were found by other authors (Gierada et al., 1998; Testa et al., 2011; Yamaguti et al., 2007), but in different body positions (dorsal and lateral decubitus).

Table 3
Measures of the diaphragm amplitude movement in the NB and MB groups.

DA	NB group (n = 39) Mean ± SD	MB group (n = 38) Mean ± SD	p-value
TV (cm) ^b	2.02 ± 0.67	1.64 ± 0.53	<0.01*
Sniff (cm)	3.37 ± 0.72	3.14 ± 1.03	0.26
LTC (cm) ^a	3.87 ± 0.94	3.48 ± 0.89	0.03*

SD: standard deviation; DA: diaphragm amplitude movement; TV: tidal volume; Sniff: fast and short inspiration; TLC: total lung capacity.

* Significant difference.

^a T-test for independent data.

^b Mann–Whitney test.

Postural misalignment may also explain the difference observed between groups in DA. Forward head posture, frequently observed in MB, induces thorax elevation by excessive use of the SCM muscle, decreasing the diaphragm muscle effectiveness (Corrêa and Bérzin, 2008). Moreover, mouth-breathing mode may produce hypertrophy of the accessory inspiratory muscles and a disadvantage of diaphragm muscle action due to its inactivity and lack of synergism with abdominal muscles (Hruska, 1997). On the other hand, Yi et al. (2008) did not find relation between the column curves and the diaphragmatic amplitude, evaluated by videofluoroscopy. The authors identified smaller diaphragmatic amplitude in MB children, compared to the NB ones. Craniocervical posture was not assessed, which is a limitation of this study, considering it could influence the muscle function. It is suggested that this evaluation should be encompassed in further studies. Moreover, it was not considered how long the participants have presented this unusual breathing mode.

This investigation is important because, despite the absence of obstruction, the functional MB may lead to changes in ventilatory function, with lower muscle strength and exercise capacity and with negative effect in the quality of life, as found in the domain of health general status by Milanese et al. (2014).

The results of the present study suggest that reduced muscular activity may negatively reflect exercise capacity and tolerance. Thus, further studies with maximal oxygen consumption test may investigate the effect of this muscular disadvantage on oxygenation and general health of these patients.

The contribution of this study for clinical practice is related to the importance of a global investigation of breathing mode, not only involving medical, orthodontics and myofunctional intervention, but also the ventilatory and muscular aspects, by physical therapist. Currently, the evaluation of this condition is fragmented, restricted to specific aspects related to each profession. Besides, little attention is given to mouth-breathing child along their development up to adult age.

5. Conclusion

Mouth-breathing mode in adults resulted in lower recruitment of the accessory inspiratory muscles during fast inspiration, and lower amplitude of the diaphragmatic movement compared to the nasal breathing.

Conflict of interest

The authors declare that there are no conflicts of interest.

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