*Am J Physiol Gastrointest Liver Physiol* 305: G862–G867, 2013.
First published October 10, 2013; doi:10.1152/ajpgi.00054.2013.

# Inspiratory muscle training improves antireflux barrier in GERD patients

**Miguel Ângelo Nobre e Souza,[1] Maria Josire Vitorino Lima,[1] Giovanni Bezerra Martins,[2] Rivianny Arrais Nobre,[1] Marcellus Henrique Loiola Ponte Souza,[4] Ricardo Brandt de Oliveira,[3] and Armênio Aguiar dos Santos[4]**

[1]*Department of Clinical Medicine, Federal University of Ceará, Ceará, Brazil;* [2]*Department of Surgery, Federal University of Ceará, Ceará, Brazil;* [3]*Department of Clinical Medicine, Ribeirão Preto School of Medicine, University of São Paulo, São Paulo, Brazil; and* [4]*Department of Physiology and Pharmacology, Federal University of Ceará, Ceará, Brazil*

Submitted 21 February 2013; accepted in final form 7 October 2013

**Nobre e Souza MÂ, Lima MJ, Martins GB, Nobre RA, Souza MH, de Oliveira RB, dos Santos AA.** Inspiratory muscle training improves antireflux barrier in GERD patients. *Am J Physiol Gastrointest Liver Physiol* 305: G862–G867, 2013. First published October 10, 2013; doi:10.1152/ajpgi.00054.2013.—The crural diaphragm (CD) is an essential component of the esophagogastric junction (EGJ), and inspiratory exercises may modify its function. This study's goal is to verify if inspiratory muscle training (IMT) improves EGJ motility and gastroesophageal reflux (GER). Twelve GER disease [GERD; 7 males, 20–47 yr, 9 esophagitis, and 3 nonerosive reflex disease (NERD)] and 7 healthy volunteers (3 males, 20–41 yr) performed esophageal pH monitoring, manometry, and heart rate variability (HRV) studies. A 6-cm sleeve catheter measured average EGJ pressure during resting, peak inspiratory EGJ pressures during sinus arrhythmia maneuver (SAM) and inhalations under 17-, 35-, and 70-cm$H_2O$ loads (TH maneuvers), and along 1 h after a meal. GERD patients entered a 5-days-a-week IMT program. One author scored heartburn and regurgitation before and after IMT. IMT increased average EGJ pressure ($19.7 \pm 2.4$ vs. $29.5 \pm 2.1$ mmHg; $P < 0.001$) and inspiratory EGJ pressure during SAM ($89.6 \pm 7.6$ vs. $125.6 \pm 13.3$ mmHg; $P = 0.001$) and during TH maneuvers. The EGJ-pressure gain across 35- and 70-cm$H_2O$ loads was lower for GERD volunteers. The number and cumulative duration of the transient lower esophageal sphincter relaxations decreased after IMT. Proximal progression of GER decreased after IMT but not the distal acid exposure. Low-frequency power increased after IMT and the higher its increment the lower the increment of supine acid exposure. IMT decreased heartburn and regurgitation scores. In conclusion, IMT improved EGJ pressure, reduced GER proximal progression, and reduced GERD symptoms. Some GERD patients have a CD failure, and IMT may prove beneficial as a GERD add-on treatment.

crural diaphragm; GERD; lower esophageal sphincter; muscle training

GASTROESOPHAGEAL REFLUX DISEASE (GERD) is a "condition which develops when the reflux of stomach contents causes troublesome symptoms and/or complications" (28). GERD symptoms affect ~5% of the population in Asia and 15% in the Western world, comprising a significant medical condition (8).

The primary line of defense against GERD is the unity of the antireflux barrier. The crural diaphragm (CD) exerts an extrinsic sphincteric action at the esophagogastric junction (EGJ) and is a key component of the antireflux barrier (18). In GERD, the increase of EGJ inspiratory pressure is reduced, further setting up the importance of the CD (21). Most episodes of gastroesophageal reflux (GER) occur during transient lower esophageal sphincter relaxations (tLESR). Indeed, CD also relaxes during tLESR (18). The autonomic nervous system (ANS) controls the EGJ relaxation and may be impaired in GERD (6).

Since the CD is an inspiratory striated muscle, its function may be modified by training (11). In fact, inspiratory muscle training (IMT) may increase diaphragm strength and tone in different clinical settings (2, 16). There is some evidence that prolonged inspiratory effort can improve gastroesophageal reflux. In children with adenotonsillar hypertrophy and obstructive sleep apnea syndrome, there are both GERD and prolonged inspiratory effort due to airway obstruction. Despite most of such children having pathological reflux, those with the worst apnea indexes tended to have less esophageal acid exposure (20). Had such children naturally trained their diaphragms, therefore, compensating a bit for their antireflux barrier weaknesses?

The goal of this studied is to verify if IMT improves EGJ motor function, autonomic function, and GERD.

## METHODS

Twelve subjects (7 males, 20–47 yr old) with the diagnosis of GERD [8 grade A, 1 grade B, Los Angeles Classification; 3 nonerosive reflex disease (NERD) defined by ambulatory esophageal pH monitoring] were selected to participate in this study. Three GERD volunteers had a 2-cm hiatal hernia. All GERD volunteers presented heartburn and attended the Gastroenterology Outpatient Facility at Walter Cantídio University Hospital (Federal University of Ceará). The study protocol had been publicly announced at the hospital. Also, seven healthy volunteers (3 males, 20–41 yr old) without any typical or atypical GERD symptom were studied. A gastroenterologist interviewed GERD volunteers and scored the frequencies of both heartburn and regurgitation with a standardized questionnaire (0: no symptom; 1: less than once a week; 2: once a week; 3: 2–4 times a week; and 4: more than 5 times a week). All volunteers had a normal physical examination and no antecedents of abdominal surgery. Written informed consent was obtained from each participant. The Research Ethics Committee of the Walter Cantídio University Hospital approved the study protocol before the experiments (no. 044.06.09).

*EGJ manometry.* Esophageal manometry was performed after a 4-h fast using an eight-lumen catheter with a 6-cm sleeve at the distal end. There were seven side-hole recording orifices, one 1 cm distal to the sleeve, and six at 3-cm intervals, starting 3 cm proximal to the sleeve (Arndorfer Specialties, Greendale, WI). A low-compliance pneumohydraulic pump (JS Biomedicals, Ventura, CA) perfused the sleeve and the side holes with distilled water at 0.5 ml/min. The catheter was connected to external pressure transducers, which were coupled to a manometry system (Synectics Medical/Polygram, Stockholm, Sweden). The sleeve straddled the EGJ 1 cm deeper to the pressure inversion point so that deep inspiration yielded a positive pressure wave. All volunteers and patients were studied in the supine position.

*EGJ pressure measurements.* The sleeve measured the average EGJ pressure during normal respiration and no swallowing and the peak

Address for reprint requests and other correspondence: M. Â. Nobre e Souza, Dept. of Clinical Medicine, Federal Univ. of Ceará, Rua Prof. Costa Mendes 1608-4° andar CEP: 60430-040, Fortaleza, Ceará, Brazil (e-mail: mans@ufc.br).

EGJ pressures during two respiratory maneuvers. The average EGJ pressure comprises both inspiratory and expiratory pressures. All the points along a 15-s swallow-free window on the EGJ pressure tracing were averaged to yield it, referenced to intragastric pressure. The first respiratory maneuver consisted of six cycles of 5-s deep inhalation and 5-s exhalation without airflow resistance [sinus arrhythmia maneuver (SAM); Ref. 9]. SAM produced six inspiratory peak pressures of the EGJ that were averaged to yield the SAM pressure. The second maneuver consisted of a quick and forced inhalation through a device that incorporated a flow-independent one-way spring-loaded valve that provided an adjustable airflow resistance (in $cmH_2O$; Ref. 2; Threshold IMT; Philips Respironics). Each subject carried on inhalations under 17-, 35-, and 70-$cmH_2O$ resistance loads and the inspiratory peak pressures under each load were registered: threshold (TH) maneuvers. All maneuvers were carried out twice and the inspiratory peak pressures of each maneuver were presented as averages of the respective individual pressures. Inspiratory pressures of the respiratory maneuvers were referenced to the mean EGJ pressure during a stable 30-s period before the maneuvers.

*Assessment of tLESR.* After the respiratory maneuvers, the volunteers drank a 200-ml chocolate-soya liquid meal (117 kcal, 16 g carbohydrates, 5.6 g protein, and 3.3 g fat; ADES, Unilever, Brazil) and lay down in the right lateral position. The sleeve position was fine tuned so that it could measure the highest postprandial LES pressure, which was registered for 1 h. tLESRs were analyzed blindly by one of the authors (M. A. Nobre e Souza) according to Holloway's criteria. All relaxations $>10$ s irrespective of swallowing were considered tLSER if they fulfilled the following criteria: relaxation rate of $\geq 1$ mmHg/s, time from onset to complete relaxation of $\leq 10$ s, and nadir pressure of $\leq 2$ mmHg (14). All relaxations were longer than 13 s.

*Esophageal pH monitoring.* Ambulatory 24-h esophageal pH was monitored using a probe with two antimony sensors 15 cm apart, with an external skin reference, in 11 GERD volunteers, before and after IMT (Alacer Biomédica). Data was stored on a single portable digital recorder (AL-3; Alacer Biomédica). Before each study, the pH probes were calibrated in buffer solutions of pH 7 and pH 1. The distal pH sensor were placed 5 cm above the proximal LES border. Volunteers reported meals, supine position, and symptoms on a diary card. Patients stopped antisecretory or prokinetic drugs at least 1 wk before the first pH study. Distal reflux was defined as a pH drop below 4. Proximal reflux was evaluated manually and was defined as drops of at least one pH unit, associated with distal reflux (29). The numbers of proximal and distal refluxes were counted for the upright, supine, and total times. Distal refluxes not associated with proximal progression were also figured out.

*Autonomic function: heart rate variability.* A peripheral-lead electrocardiogram (ECG) was recorded for 60 min, at 0.25 Hz (WinCardio; Micromed Biotecnologia), after a 15-min adaptation period. The ECG signal was analog-to-digital converted with a sampling rate of 200 Hz. The signal was recorded, and the appropriate time series of RR intervals were generated. The R peak was determined by a maximum of the peak value. One operator (M. A. Nobre e Souza) revised all ECG signals manually and chose a 30-min period free of artifact or ectopic beats. This period was used to compute frequency domain components of spectral heart rate variability (HRV). The mean number of analyzed R peaks was 196 ± 34. HRV was analyzed in the frequency domain with a MATLAB-based software (Kubios HRV Software, Finland). A power spectrum density (PSD) estimate was calculated for the RR interval series. The regular PSD estimators implicitly assume equidistant sampling, and thus the RR interval series was converted to equidistantly sampled series by cubic spline interpolation before PSD estimation. The HRV spectrum was calculated with the FFT-based Welch's periodogram method. Very low frequency (VLF, 0–0.04 Hz), low frequency (LF, 0.04–0.15 Hz), high frequency (HF, 0.15–0.4 Hz), and total powers (T) were figured out in squared milliseconds ($ms^2$; Ref. 1).

*IMT program.* After the initial manometric, pH, and autonomic functional studies, GERD volunteers enrolled in an inspiratory 5-days-a-week IMT program under progressive inspiratory resistance. A physical therapist (M. J. V. Lima) managed the exercise program held in the Walter Cantídio University Hospital outpatient facility. Initial resistance was set at 30% of maximal inspiratory pressure (maxIP) and was increased, as long as tolerated, by 5% every 5 days for 2 mo. Each IMT session consisted of 10 series of 15 inspirations and lasted ~30 min (10). maxIP was measured with an analog vacuumeter, and the inspiratory resistance was accomplished with the Threshold IMT device described previously in the text. At some point during the IMT, two Threshold IMT devices connected in series were generally needed to accomplish the required inspiratory resistance. All initial functional studies were repeated within 1 wk of the IMT end. One volunteer did not perform the pH and autonomic studies at the end of the protocol.

*Statistical analysis.* The number of proximal reflux progression was presented as the difference between the number of nonprogressing distal reflux and proximal reflux. The increment of acid exposure (%pH <4) and LF power after IMT were tested for correlation (Spearman's rank correlation). Scores, number of tLESR, and number of GER that progressed proximally were presented in median and range. Continuous data are presented as means ± SE. Student's paired *t*-test was employed to compare quantitative and continuous variables before and after IMT and unpaired *t*-test to compare healthy and GERD volunteer variables. Wilcoxon's matched pair test was used to compare the distribution of quantitative and discrete variables before and after IMT. The EGJ pressure difference between 70 and 35 $cmH_2O$ inspiratory loads was compared between healthy and GERD volunteers. The level of statistical significance was set at 0.05 for differences in mean values and distributions [JMP Statistical Discovery Software, version 7.0.1, SAS Institute (Cary, NC); GraphPad Prism, GraphPad Software (La Jolla, CA)].

## RESULTS

All GERD volunteers presented heartburn at least once a week, and 10 also had regurgitation. The scores of heartburn and regurgitation decreased significantly after IMT [3 (3–4) vs. 0 (0–0.7), $P = 0.003$; and 2.5 (1–3.7) vs. 0 (0–0), $P = 0.008$, respectively].

Average EGJ pressure in GERD volunteers (19.7 ± 2.4 mmHg) was similar to healthy ones (25.5 ± 5.6 mmHg, $P = 0.256$) and increased after IMT (29.5 ± 2.1 mmHg, $P < 0.001$; Fig. 1). Inspiratory EGJ pressure during SAM was nonsignificantly lower in GERD volunteers (89.6 ± 7.6 mmHg) than in

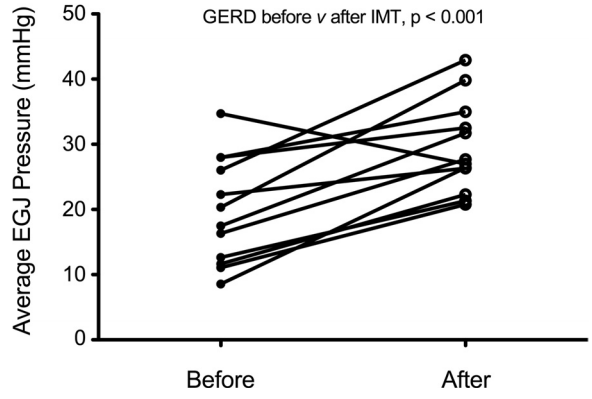


Fig. 1. Average resting esophagogastric junction (EGJ) pressure in gastroesophageal reflux disease (GERD) volunteers increased significantly after inspiratory muscle training (IMT). EGJ pressure was the average of inspiratory and expiratory pressures during a 15-s swallow-free period (paired *t*-test).

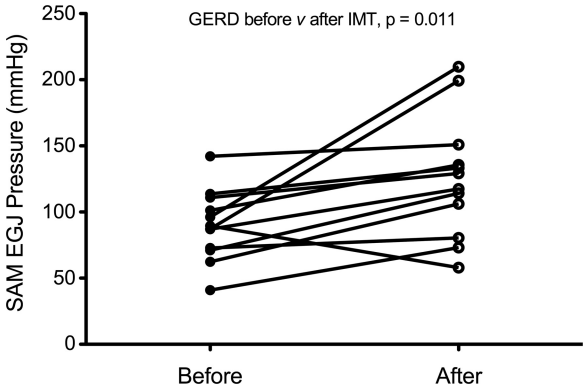


Fig. 2. Inspiratory EGJ pressure during sinus arrhythmia maneuver (SAM) in GERD volunteers increased significantly after IMT. EGJ pressure was measured at the end of deep 5-s inhalations (paired *t*-test).

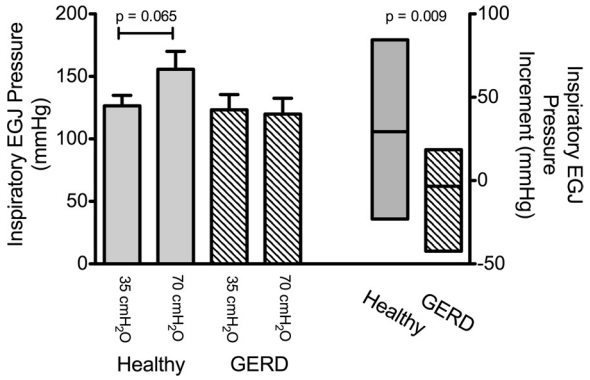


Fig. 3. Increasing inspiratory load from 35 to 70 $cmH_2O$ yielded a greater inspiratory EGJ pressure in the healthy group but not in the GERD one (bars at *left*). The increment in inspiratory EGJ pressure across the 35- and the 70-$cmH_2O$ loads was significantly higher for the healthy group (floating bars at *right*). Data are means ± SE at the bars (paired *t*-test) and minimum, maximum, and mean at the floating bars (unpaired *t*-test).

healthy ones (119.4 ± 14.4 mmHg, $P = 0.06$) and increased significantly after IMT (125.6 ± 13.3 mmHg, $P = 0.001$; Fig. 2). Inspiratory EGJ pressures during inspiratory loads of 17, 35, and 70 $cmH_2O$ increased significantly after IMT (Table 1). Increasing inspiratory load from 35 to 70 $cmH_2O$ yielded a greater inspiratory EGJ pressure in the healthy group (120.2 ± 6.5 vs. 154 ± 16.4 mmHg, $P = 0.065$) but not in the GERD one (123.3 ± 12.2 vs. 119.9 ± 12.6 mmHg, $P = 0.494$; Fig. 3). The increment in EGJ inspiratory pressure between the 70- and the 35-$cmH_2O$ loads was significantly higher for the healthy group (33.8 ± 14.6 vs. −3.4 ± 4.8 mmHg, $P = 0.009$; Fig. 3). The inspiratory load of 17 $cmH_2O$ yielded an inspiratory EGJ pressure similar to the 35-$cmH_2O$ load in healthy volunteers (137.3 ± 16.2 mmHg, $P = 0.193$, vs. 35 $cmH_2O$) and lower in GERD ones (110.9 ± 11.5 mmHg, $P = 0.009$, vs. 35 $cmH_2O$).

The number of tLESR in GERD volunteers decreased after IMT [8.5 events/h (4–17) vs. 7 events/h (2–13), $P = 0.032$]. The sum of tLESR durations was shorter after IMT (199.1 ± 23.5 vs. 156.8 ± 25.9 min, $P = 0.034$). The mean durations of the tLESR events were similar before and after IMT (22 ± 1.3 vs. 21.7 ± 1.3 min, $P = 0.91$).

Total acid exposure before and after IMT was similar, both for the proximal (10.4 ± 4.4 vs. 12.5 ± 4.1 min, respectively, $P = 0.751$) and distal (50.9 ± 15.1 vs. 56.9 ± 13.1 min, respectively, $P = 0.765$) esophagus. Proximal progression of reflux was lower after IMT [−8 (−16; 5) vs. −10 (−28; −3), $P = 0.04$] (Fig. 4). Such a phenomenon was mostly due to a reduced proximal progression of upright reflux [−8 (−19; 5) vs. −14 (−30; −3), $P = 0.041$]. Supine reflux did not progress significantly less after IMT [−21 (−30; −12) vs. −19 (−42; −11), $P = 0.26$].

The LF band power of HRV increased after IMT (Table 2). The difference of the LF power after and before IMT correlated negatively with the difference of supine acid exposure after and before the workout. The higher the increment in LF power, the lower the increment of supine acid exposure (%time pH <4) after IMT, both in the proximal ($r = -0.615$, $P = 0.044$) and distal esophagus ($r = -0.755$, $P = 0.007$; Table 3).

Table 1. *Inspiratory EGJ pressures across inspiratory loads of 17, 35, and 70 $cmH_2O$ increased significantly after IMT*

| Inspiratory Load | Healthy Volunteers ($n = 7$): EGJ pressure (No IMT) | GERD Volunteers ($n = 12$): EGJ Pressure (IMT) | | |
|---|---|---|---|---|
| | | Before | After | *P* |
| 17 $cmH_2O$ | 137.3 ± 16.2 | 110.9 ± 11.5 | 141 ± 11.3 | 0.002 |
| 35 $cmH_2O$ | 120.2 ± 6.5 | 123.3 ± 12.2 | 148.6 ± 10.6 | 0.015 |
| 70 $cmH_2O$ | 154 ± 16.4 | 119.9 ± 12.6 | 149.2 ± 10.1 | 0.008 |

Values are means ± SE. IMT, inspiratory muscle training; GERD, gastroesophageal reflux disease; EGJ, esophagogastric junction. *P* values before vs. after IMT by paired *t*-test.

## DISCUSSION

The main results of our study show that both average and inspiratory EGJ pressures are increased, and tLESR rate, the proximal esophagus acid exposure, as well as the GER symptoms are reduced in GERD patients by IMT. Also, there is a graded increment in inspiratory pressure during TH maneuvers in healthy controls but not in GERD patients. Since CD phasic activity contributes to EGJ pressure (18), the increase in inspiratory EGJ pressure is probably a direct consequence of an IMT-induced enhancement of CD strength. Similarly, inspiratory muscle strengthening by IMT has been well described in respiratory diseases (5). Since the average EGJ pressure consisted of both inspiratory and expiratory pressures, its increase after IMT was probably due to the increase in inspiratory EGJ

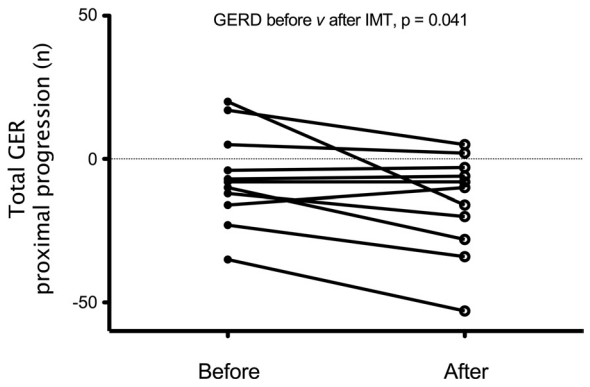


Fig. 4. Total GER proximal progression decreased significantly after IMT. Data presented as the difference between the number of proximal reflux and nonprogressing reflux (paired *t*-test).

Table 2. *Heart rate variability (LF power) increased after IMT*

| | Before | After | *P* |
|---|---|---|---|
| VLF | 1,942 ± 357.6 | 3806 ± 1157 | 0.068 |
| LF | 363 ± 69.3 | 511.4 ± 112.1 | 0.038 |
| HF | 268.6 ± 44.2 | 216.5 ± 34.4 | 0.248 |
| Total | 2,573 ± 435.4 | 4,534 ± 1266 | 0.070 |
| LF/HF | 1,332 ± 391.8 | 2,273 ± 516.6 | 0.041 |

Values are means ± SE (paired *t*-test); $n = 11$. VLF, very low frequency; LF, low frequency; HF, high frequency.

pressure. Moreover, CD tone may also have changed after IMT. Striated muscle tone depends physiologically on two factors: the basic viscoelastic properties of the soft tissues associated with the muscle and the degree of activation of the contractile apparatus of the muscle (26). IMT would change the contractile apparatus of the CD so as to shift its length-tension relationship and increase CD tone and the average EGJ pressure.

An interesting finding in our study was the reduction of acid exposure of the proximal esophagus after IMT. GERD patients particularly with hiatal hernia have a wide, highly compliant EGJ at low-pressure distension as well as a less asymmetrical EGJ relative to healthy controls. This asymmetry probably is related to the anatomical disposition of the CD around the EGJ that seems to compress its lateral aspects. IMT may partly restore this normal asymmetry and reduce the EGJ opening by improving CD tone. If this is the case, transphincteric flow would be reduced as well as the refluxate volume (22). Consequently, migration of reflux from distal to proximal esophagus and associated symptoms would be reduced. This notion is supported by the finding that reducing the EGJ compliance by the endoscopic insertion of a hydrogel expandable prostheses diminishes the proximal progression of GER and reduces symptoms but does not modify the distal esophageal acid exposure (4). CD strengthening may improve the EGJ gatekeeper role and would decrease the number of proximal refluxes.

Smooth muscle tone and tLESR are under ANS control (13). The relationship between HRV and autonomic function is a complex phenomenon, and it is generally accepted that the greater the HRV, the healthier the individual (1). Ultimately, IMT is a physical exercise that would improve autonomic function, particularly vagal tone. IMT improves ANS function (27), similarly to regular physical training (17). IMT would drive a new and healthier balance in ANS activity that could be associated with a better EGJ motor function comprising both enhancement of the pressure generated by the smooth muscle component of LES and the decrease in the rate of tLESR. This autonomic improvement may act not only in the EGJ but also in the stomach to reduce tLESR. Disturbed postprandial meal distribution in the stomach is associated with increased tLESR in children fed with high volume and osmolality meal (25), and GERD patients have reduced proximal stomach meal retention (12). It is possible that autonomic function improvement in GERD would counteract motor derangements like the last one.

tLESR occurred at a slightly higher than expected rate in this study. It is known that chocolate results in a decrease of LES pressure and a significant increase in esophageal acid exposure (19). The high chocolate content of the test meal used here may have contributed to this tLESR rate.

Interestingly, increasing inspiratory loads across 35 and 70 $cmH_2O$ built up the EGJ pressure during forced inspiration in the healthy volunteers. This phenomenon did not occur in the GERD patients. Large hiatal hernias would explain this result, but it was not the case of our volunteers. Also, a closed glottis is associated with diaphragm relaxation and could explain the failure of GERD patients to cope with graded increase in inspiratory resistance. However, this was not the case since there was airflow to tidal volume during the TH maneuvers. There could be a failure of the CD contractile system instead. Studying inspiratory EGJ pressure during inhalations through a graded resistive device proved important in this work. Perhaps, some GERD patients may have a crural insufficiency that this maneuver unveiled.

Heartburn and regurgitation improved drastically. A placebo-like effect would explain part of these results. However, a reduction in hypersensitivity might also play a role in this phenomenon. GERD is frequently associated with hypersensitivity (15). Physical activity enhances the parasympathetic autonomic system (17), and this is associated with a reduction in hypersensitivity (3).

EGJ pressures at rest and during respiratory maneuvers were recorded with a forward perfused sleeve sensor carefully positioned so that inhalation yielded a positive pressure wave. There may be some concern about the fidelity of the sleeve sensor to accurately record inspiratory EGJ pressures. However, the two following aspects assure that this sensor was adequate for our purposes. Our experimental design was able to demonstrate an increase in inspiratory pressure in GERD patients after IMT and a graded increment in inspiratory pressure during TH maneuvers in healthy controls. Therefore, the failure of GERD patients to achieve such a graded increment in inspiratory pressure should be considered a true phenomenon. We also recorded tLESR and esophageal acid exposure. In the first case, a sleeve or a high-resolution perfused catheter seems more appropriate since solid-state probes may

Table 3. *Higher the increment in LF power the lower the increment of supine acid exposure after IMT*

| | VLF | LF | HF | Total |
|---|---|---|---|---|
| Proximal reflux | | | | |
| GER (*n*) | −0.543; 0.085 | −0.667; 0.025 | −0.051; 0.883 | −0.570; 0.067 |
| %Time <4 | −0.450; 0.165 | −0.615; 0.044 | −0.193; 0.570 | −0.495; 0.121 |
| Distal reflux | | | | |
| GER (*n*) | −0.519; 0.102 | −0.733; 0.010 | −0.132; 0.699 | −0.601; 0.050 |
| %Time <4 | −0.655; 0.029 | −0.755; 0.007 | 0.018; 0.958 | −0.718; 0.013 |

Data are Spearman's *r* and *P* values; $n = 11$. This was mainly shown by the negative correlation between the difference of heart rate variability (LF power) after and before IMT and the difference of supine acid exposure after and before the workout.

show a signal drift after several minutes of study (23). In the case of acid exposure, impedance/pH esophageal monitoring would have improved GER detection. However, a double sensor pH study was sufficient to detect proximal GER progression and to study the correlation with autonomic function variables in our conditions.

There may also be significant concerns about the practicality of the IMT program since it was intense and held in a clinical setting. A general physical exercise program for GERD patients could include IMT. Together with other GERD treatment regimens, this would be a reasonable and affordable way to treat GERD. In fact, most volunteers felt it could be done at home and inspiratory muscle trainers are not much expensive. The ones used in this work cost around $50 plus tax and were reused by the same volunteer all along the training program.

Recently, Eherer and coworkers (7) showed that vocal training reduced GERD symptoms and esophageal acid exposure. However, there was not any improvement in either the antireflux barrier or tLESR rate. This fact could be due to the lack of inspiratory load during their vocal training program. Vocal training involves changing from thoracic to abdominal breathing and would not strengthen the CD. The acid pocket position is important in the pathogenesis of GERD and can be changed pharmacologically (24). Would vocal training displace the acid pocket distally? If so, it would change GER pattern without impacting on antireflux barrier or autonomic function.

The new information presented here shows a CD failure in GERD patients and may stimulate GERD treatment trials concerning which patients would mostly benefit from IMT and which training regimen would be the most effective.

## GRANTS

This study was funded in full by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) Grant 481098/2009-7.

## DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

## AUTHOR CONTRIBUTIONS

Author contributions: M.A.N.e.S. conception and design of research; M.A.N.e.S., M.J.V.L., G.B.M., and R.A.N. performed experiments; M.A.N.e.S. analyzed data; M.A.N.e.S., G.B.M., R.A.N., M.H.L.P.S., R.B.d.O., and A.A.d.S. interpreted results of experiments; M.A.N.e.S. prepared figures; M.A.N.e.S., M.J.V.L., G.B.M., R.A.N., M.H.L.P.S., R.B.d.O., and A.A.d.S. drafted manuscript; M.A.N.e.S., M.H.L.P.S., R.B.d.O., and A.A.d.S. edited and revised manuscript; M.A.N.e.S. and R.B.d.O. approved final version of manuscript.

## REFERENCES

1. **Anonymous.** Heart rate variability: standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology/The North American Society of Pacing Electrophysiology. *Circulation* 93: 1043–1065, 1996.
2. **Bailey SJ, Romer LM, Kelly J, Wilkerson DP, DiMenna FJ, Jones AM.** Inspiratory muscle training enhances pulmonary $O_2$ uptake kinetics and high-intensity exercise tolerance in humans. *J Appl Physiol* 109: 457–468, 2010.
3. **Botha C, Knowles C, Aziz Q.** The effect of psychophysiological autonomic modulation on human esophageal pain hypersensitivity. *Neurogastroenterol Motil* 24: 1, 2012.
4. **Cicala M, Gabbrielli A, Emerenziani S, Guarino MP, Ribolsi M, Caviglia R, Costamagna G.** Effect of endoscopic augmentation of the lower oesophageal sphincter (Gatekeeper reflux repair system) on intraoesophageal dynamic characteristics of acid reflux. *Gut* 54: 183–186, 2005.
5. **Decramer M.** Response of the respiratory muscles to rehabilitation in COPD. *J Appl Physiol* 107: 971–976, 2009.
6. **Dobrek L, Nowakowski M, Mazur M, Herman RM, Thor PJ.** Disturbances of the parasympathetic branch of the autonomic nervous system in patients with gastroesophageal reflux disease (GERD) estimated by short-term heart rate variability recordings. *J Physiol Pharmacol* 55, *Suppl* 2: 77–90, 2004.
7. **Eherer AJ, Netolitzky F, Hogenauer C, Puschnig G, Hinterleitner TA, Scheidl S, Kraxner W, Krejs GJ, Hoffmann KM.** Positive effect of abdominal breathing exercise on gastroesophageal reflux disease: a randomized, controlled study. *Am J Gastroenterol* 107: 372–378, 2012.
8. **El-Serag HB.** Time trends of gastroesophageal reflux disease: a systematic review. *Clin Gastroenterol Hepatol* 5: 17–26, 2007.
9. **Ewing DJ, Martyn CN, Young RJ, Clarke BF.** The value of cardiovascular autonomic function tests: 10 years experience in diabetes. *Diabetes Care* 8: 491–498, 1985.
10. **Geddes EL, Reid WD, Crowe J, O'Brien K, Brooks D.** Inspiratory muscle training in adults with chronic obstructive pulmonary disease: a systematic review. *Respir Med* 99: 1440–1458, 2005.
11. **Guyton AC, Hall JE.** *Guyton and Hall Textbook of Medical Physiology*. Philadelphia, PA: Saunders, 2011.
12. **Herculano JR Jr, Troncon LE, Aprile LR, Moraes ER, Secaf M, Onofre PH, Dantas RO, Oliveira RB.** Diminished retention of food in the proximal stomach correlates with increased acidic reflux in patients with gastroesophageal reflux disease and dyspeptic symptoms. *Dig Dis Sci* 49: 750–756, 2004.
13. **Holloway RH.** Systemic pharmacomodulation of transient lower esophageal sphincter relaxations. *Am J Med* 111 *Suppl 8A*: 178S–185S, 2001.
14. **Holloway RH, Penagini R, Ireland AC.** Criteria for objective definition of transient lower esophageal sphincter relaxation. *Am J Physiol Gastrointest Liver Physiol* 268: G128–G133, 1995.
15. **Knowles CH, Aziz Q.** Visceral hypersensitivity in non-erosive reflux disease. *Gut* 57: 674–683, 2008.
16. **Kodric M, Trevisan R, Torregiani C, Cifaldi R, Longo C, Cantarutti F, Confalonieri M.** Inspiratory muscle training for diaphragm dysfunction after cardiac surgery. *J Thorac Cardiovasc Surg* 145: 819–823, 2013.
17. **Levy WC, Cerqueira MD, Harp GD, Johannessen KA, Abrass IB, Schwartz RS, Stratton JR.** Effect of endurance exercise training on heart rate variability at rest in healthy young and older men. *Am J Cardiol* 82: 1236–1241, 1998.
18. **Mittal RK, Rochester DF, McCallum RW.** Electrical and mechanical activity in the human lower esophageal sphincter during diaphragmatic contraction. *J Clin Invest* 81: 1182–1189, 1988.
19. **Murphy DW, Castell DO.** Chocolate and heartburn: evidence of increased esophageal acid exposure after chocolate ingestion. *Am J Gastroenterol* 83: 633–636, 1988.
20. **Noronha AC, de Bruin VM, Nobre e Souza MA, de Freitas MR, Araujo Rde P, Mota RM, de Bruin PF.** Gastroesophageal reflux and obstructive sleep apnea in childhood. *Int J Pediatr Otorhinolaryngol* 73: 383–389, 2009.
21. **Pandolfino JE, Kim H, Ghosh SK, Clarke JO, Zhang Q, Kahrilas PJ.** High-resolution manometry of the EGJ: an analysis of crural diaphragm function in GERD. *Am J Gastroenterol* 102: 1056–1063, 2007.
22. **Pandolfino JE, Shi G, Trueworthy B, Kahrilas PJ.** Esophagogastric junction opening during relaxation distinguishes nonhernia reflux patients, hernia patients, and normal subjects. *Gastroenterology* 125: 1018–1024, 2003.
23. **Robertson EV, Lee YY, Derakhshan MH, Wirz AA, Whiting JR, Seenan JP, Connolly P, McColl KE.** High-resolution esophageal manometry: addressing thermal drift of the manoscan system. *Neurogastroenterol Motil* 24: 61–64, e11, 2012.
24. **Rohof WO, Bennink RJ, de Ruigh AA, Hirsch DP, Zwinderman AH, Boeckxstaens GE.** Effect of azithromycin on acid reflux, hiatus hernia and proximal acid pocket in the postprandial period. *Gut* 61: 1670–1677, 2012.
25. **Salvia G, De Vizia B, Manguso F, Iula VD, Terrin G, Spadaro R, Russo G, Cucchiara S.** Effect of intragastric volume and osmolality on mechanisms of gastroesophageal reflux in children with gastroesophageal reflux disease. *Am J Gastroenterol* 96: 1725–1732, 2001.
26. **Simons DG, Mense S.** Understanding and measurement of muscle tone as related to clinical muscle pain. *Pain* 75: 1–17, 1998.

27. **Souza MA, Lima, MJ, Gomes TN, Souza MH, Santos AA.** Inspiratory diaphragm workout increases heart rate variability and improves GERD symptoms. *Gastroenterology, Suppl* 1 140: S-204, 2011.
28. **Vakil N, van Zanten SV, Kahrilas P, Dent J, Jones R; Global Consensus Group.** The Montreal definition and classification of gastroesophageal reflux disease: a global evidence-based consensus. *Am J Gastroenterol* 101: 1900–1920; quiz 1943, 2006.
29. **Wolf C, Timmer R, Breumelhof R, Seldenrijk CA, Smout AJ.** Prolonged measurement of lower oesophageal sphincter function in patients with intestinal metaplasia at the oesophagogastric junction. *Gut* 49: 354–358, 2001.