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Effects of Controlled Breathing, Mental Activity and Mental Stress With or Without Verbalization on Heart Rate Variability

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OBJECTIVES To assess whether talking or reading (silently or aloud) could affect heart rate variability (HRV) and to what extent these changes require a simultaneous recording of respiratory activity to be correctly interpreted.

BACKGROUND Sympathetic predominance in the power spectrum obtained from short- and long-term HRV recordings predicts a poor prognosis in a number of cardiac diseases. Heart rate variability is often recorded without measuring respiration; slow breaths might artefactually increase low frequency power in RR interval (RR) and falsely mimic sympathetic activation.

METHODS In 12 healthy volunteers we evaluated the effect of free talking and reading, silently and aloud, on respiration, RR and blood pressure (BP). We also compared spontaneous breathing to controlled breathing and mental arithmetic, silent or aloud. The power in the so called low- (LF) and high-frequency (HF) bands in RR and BP was obtained from autoregressive power spectrum analysis.

RESULTS Compared with spontaneous breathing, reading silently increased the speed of breathing ($p < 0.05$), decreased mean RR and RR variability and increased BP. Reading aloud, free talking and mental arithmetic aloud shifted the respiratory frequency into the LF band, thus increasing LF% and decreasing HF% to a similar degree in both RR and respiration, with decrease in mean RR but with minor differences in crude RR variability.

CONCLUSIONS Simple mental and verbal activities markedly affect HRV through changes in respiratory frequency. This possibility should be taken into account when analyzing HRV without simultaneous acquisition and analysis of respiration. (J Am Coll Cardiol 2000;35: 1462–9) © 2000 by the American College of Cardiology

Heart rate variability (HRV) is of increasing interest to clinicians after the demonstration that low variability is associated with a poor long-term prognosis after myocardial infarction or heart failure (1,2). Furthermore, this prognostic information is in some cases additive to the usual obvious clinical predictors such as left ventricular ejection fraction. A recent report, mostly based on the analysis of HRV from Holter recordings suggested practical guidelines on how to carry out satisfactory HRV measurements (3). However, under the conditions typically found in Holter recordings (i.e., subjects free to engage in normal activities), HRV can be influenced by other nonpathological conditions such as activity (4) or different patterns of breathing (5,6). Despite this evidence that slow breaths can generate slow fluctuations that can be confused with those related to sympathetic activity, respiration is generally not measured in Holter studies of long-term HRV. To our knowledge there have been no studies comprehensively comparing the effects of controlled breathing (i.e., pure changes in respiration) or of mental arithmetic (i.e., pure change in arousal) on HRV or of the different types of verbal activities with slow respiratory rates (simple free talking, reading aloud and mental arithmetic aloud) or of comparison with similar activities performed silently (spontaneous breathing, controlled breathing, reading silently and silent mental arithmetic), despite the obvious fact that these are common activities that clearly alter breathing patterns and arousal.

We, therefore, investigated whether talking or reading (silently or aloud) could in themselves lead to effects on HRV and to what extent this mental activity could change the power spectrum and mimic sympathetic activation. We measured HRV, respiration and blood pressure (BP) in a group of healthy subjects during controlled breathing (i.e., absence of verbal respiratory changes and stressful mental

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Abbreviations and Acronyms

BP = blood pressure
HF = high frequency
HRV = heart rate variability
LF = low frequency
nu = normalized units
RR = RR interval
SEM = standard error of the mean

activity), reading aloud, reading silently and mental arithmetic (multiple subtractions performed either silently or aloud), in order to ascertain the interaction between changes in respiratory pattern, mental arousal, verbalization and HRV. We studied controlled breathing in order to eliminate the possibility that occasional slower or faster breaths could superimpose on the heart rate spectra at the frequencies of these irregular breaths. Since reading aloud would clearly impose a different respiratory pattern than normal breathing, we compared this with reading silently. Since reading might be regarded as a mild arousal, and since arousal is known to depress the gain of the baroreflex control of the sinus node (7), we also studied the effect of definite stress by performing mental arithmetic both silently and aloud.

METHODS

The study was performed in 12 normotensive male volunteers (average age 29 ± 1 years, mean ± SEM). None of the subjects was taking medication. The subjects gave written informed consent and the study protocol was approved by the Ethics Committee of the University of Pavia, Italy.

Experimental procedure. All the tests were carried out supine. After 20 to 30 min, rest and familiarization with the laboratory, recordings were made in random order during: a) spontaneous breathing (4 min); b) reading a text silently (the material was unfamiliar to the subjects and "neutral"; the text was held in front of the subject) (3 min); c) reading the same text aloud (3 min); d) normal talking (the subject reporting his usual daily activities, 3 min); e) mental stress without talking: the subject was repeatedly subtracting 7 from nonrecurring figures under time pressure while writing the results on a blackboard held in front of the subject (3 min); f) the same tests aloud (3 min). Finally, at the end of the protocol the following recordings were obtained:

1) controlled breathing at 0.25 Hz (i.e., 15 breaths/min for 3 min);
2) controlled breathing at a respiratory frequency similar to that occurring spontaneously during silent reading (b) with spontaneous breathing (for 3 min). These recordings were performed last, in order not to influence the subject's breathing pattern during the other interventions (controlled breathing) and in order to have an estimate of the frequency of breathing during silent reading.

Data acquisition. Simultaneous recordings of electrocardiogram, respiration (obtained from an inductive pneumograph) and noninvasive BP (Finapres, Ohmeda, Englewood, Colorado) were digitized on-line at a sampling rate of 250 sample/s for each channel by means of a multifunction hardware and software (8) and simultaneously stored on hard disk. From these signals we obtained the synchronized time series of RR interval (RR), respiration and BP. The pneumogram was sampled at the peak of each R wave and was expressed in arbitrary units (a.u.). The pneumogram bears—for each subject—a linear relationship with tidal volume, so that relative changes in minute ventilation during the various interventions could be compared for each subject (9). Because the goal of the study was to study the relationship between breathing pattern and verbal or mental activities, we only measured relative changes in minute ventilation and did not attempt to calibrate the pneumogram in absolute units.

Data analysis. We applied power spectral analysis to RR and respiration using an autoregressive model (10). Unlike other methods of computing the power spectrum (as for example, the fast Fourier transform), the autoregressive method has the advantage of giving reliable estimates of the power associated with the peaks at various frequencies using a relatively small amount of data. In addition, it is able to identify the frequency of each significant peak (10). Spectral components were obtained by a decomposition method to measure the area below each spectral peak (10). The respiration-related oscillations of the RR were identified by comparison with the oscillations of the respiratory spectrum. Spectral analysis of the RR showed two main components: a frequency component between 0.03 to 0.14 Hz (low-frequency peak [LF]) and a second component of RR at a higher frequency (over 0.15 Hz, high-frequency [HF]). In general, the low-frequency component has been thought to reflect mainly the sympathetic nervous activity both at the cardiac (with vagal influences) and vascular level (3,11), while the HF, respiratory component reflects vagal activity on the heart together with nonneural changes, which follow inspiration induced increases in venous return (5,8). However, since a variable respiratory pattern was the focus of this study, no assumption a priori was made about the significance of the LF component in the RR; rather we analyzed the dependence of LF on breathing during the various interventions.

Statistics. Values are presented as mean ± standard error of the mean (SEM). Due to their skewed distribution, the power in the LF and HF bands were analyzed statistically only after natural logarithmic transformation. In addition the relative (percent) proportion of LF and HF components in the RR and respiration were expressed as percentages of the power comprised between 0.03 and 0.40 Hz (normalized units, nu). Statistical analysis was performed using repeated measures—analysis of variance for repeated measures on two factors, to establish the relative contribution of

reading and verbalization, or of verbalization and stress, on the respiratory and RR data. Pairwise comparisons were made using the Scheffé test. Probability values of <0.05 were considered statistically significant.

RESULTS

Complete results are shown in Tables 1 (RR) and 2 (BP). Figure 1 illustrates the respiratory changes that occurred in the different protocol conditions.

Effect of controlled versus free breathing. *RR interval.* Controlled breathing at either 15 breaths per min or at a frequency similar to that of silent reading (i.e., ~18 breaths per min) did not change RR mean value and variability and increased the power of the respiratory fluctuations (in relative percentage terms). The power in the LF band was not significantly reduced during controlled breathing.

Respiration. Compared with spontaneous breathing, controlled breathing increased ventilation by 131.9 ± 36.5% (15 breaths per min, $p < 0.01$) and by 192.2 ± 46.9% (~18 breaths per min); the power in the HF band of respiration increased significantly during the slower, but not during the faster, controlled breathing, whereas the respiratory frequency increased only during the faster controlled breathing. Therefore, the increase in ventilation observed during controlled breathing was mainly due to an increase in tidal volume during slower controlled breathing and mainly to an increase in breathing frequency during faster controlled breathing.

Blood pressure. No changes in BP were observed during controlled breathing.

Overall these results indicate that controlled breathing increases the HF mainly because of an increase in ventilation; the lack of increase in mean RR suggests that controlled breathing at these frequencies might not increase vagal activity.

Effect of free talking, reading aloud versus silent reading and versus baseline control (spontaneous breathing). *RR interval.* Compared with spontaneous breathing, both reading (silently and aloud) and free talking decreased RR (aloud more than silently), whereas the RR variability decreased only during reading silently, but tended to increase during free talking (although it did not reach statistical significance). Also the power in the LF band tended to increase during all conditions associated with verbalization (reading aloud and free talking) although it increased significantly only during the latter (Table 1). The power in the HF component decreased only during reading silently. In relative terms, we have observed a tendency toward an increase in LF nu during all conditions, although a significant increase occurred only during free talking compared with quiet spontaneous breathing (Fig. 2). When we compared reading silently with no reading but at a similar respiratory rate (faster controlled breathing ~18 breaths per min), we observed during silent reading a reduced RR

Table 1. RR Interval Power Spectral Analysis Results (Mean ± SEM)

	Spontaneous Breathing	Controlled Breathing (15 breaths/min)	Controlled Breathing (18 breaths/min)	Silent Reading	Reading Aloud	Free Talking	Mental Stress Aloud	Mental Stress Silent
Mean (ms)	920 ± 20	907 ± 25	937 ± 18†§§¶¶¶###&&&	878 ± 22**¶¶¶	825 ± 24**†††###¶¶¶	809 ± 22**‡‡¶	769 ± 27***	724 ± 28***#
Standard deviation (ms)	56.85 ± 6.82	57.80 ± 12.04	47.72 ± 10.36§§	41.39 ± 5.18**##	59.46 ± 6.50††	71.62 ± 13.93	63.84 ± 5.85*	44.72 ± 5.02##
LFpw (ln-ms^2)	6.21 ± 0.67	6.18 ± 0.38	5.58 ± 0.41†§§§###&&&	6.24 ± 0.39#	7.09 ± 0.33‡	7.65 ± 0.38*‡‡‡¶	7.21 ± 0.23	5.74 ± 0.58#
HFpw (ln-ms^2)	6.23 ± 0.45	6.76 ± 0.51	5.55 ± 0.47*¶&	5.76 ± 0.39*‡‡¶¶	6.56 ± 0.46#¶¶¶	5.97 ± 0.73	5.72 ± 0.45	4.93 ± 0.43***#
LFnu (%)	54.04 ± 7.23	39.54 ± 8.81	40.99 ± 8.56†§§§¶##	57.01 ± 5.71‡	58.99 ± 4.67‡	75.16 ± 8.11*‡‡	69.07 ± 6.07‡‡	71.05 ± 6.94‡‡
HFnu (%)	39.04 ± 5.76	57.60 ± 8.67*	37.44 ± 7.28	37.39 ± 5.91‡	37.11 ± 4.92‡	21.02 ± 7.20*‡‡	23.93 ± 5.53*‡‡‡	23.26 ± 6.85‡‡

Changes of mean RR interval, RR interval global variability and spectral components in absolute power and in relative (normalized units) terms.
HF = high frequency components; LF = low-frequency components; ln = natural logarithm; ms = milliseconds; nu = normalized units; pw = power.
*$p < 0.05$; **$p < 0.01$; ***$p < 0.001$ vs. spontaneous breathing; †$p < 0.05$; ††$p < 0.01$; †††$p < 0.001$ vs. silent reading; ‡$p < 0.05$; ‡‡$p < 0.01$; ‡‡‡$p < 0.001$ vs. controlled breathing (15 breaths/min); §$p < 0.05$; §§$p < 0.01$; §§§$p < 0.001$ vs. free talking; ¶$p < 0.05$; ¶¶$p < 0.01$; ¶¶¶$p < 0.001$ vs. mental stress; #$p < 0.05$; ##$p < 0.01$; ###$p < 0.001$ vs. mental stress aloud; &$p < 0.05$; &&$p < 0.01$; &&&$p < 0.001$ vs. reading aloud.

Table 2. Blood Pressure Results (mean ± SEM)

	Spontaneous Breathing	Controlled Breathing (15 breaths/min)	Controlled Breathing (18 breaths/min)	Silent Reading	Reading Aloud	Free Talking	Mental Stress Aloud	Mental Stress Silent
Systolic (mm Hg)	114.2 ± 2.6	110.8 ± 3.8††	112.3 ± 2.3††¶¶¶###§§&&	120.8 ± 3.0*‡‡###¶¶¶	123.7 ± 4.3*‡‡¶¶¶	121.7 ± 4.0**‡###¶¶¶	131.2 ± 4.3**‡‡‡	139.2 ± 4.5***‡‡‡#
Diastolic (mm Hg)	76.2 ± 2.4	75.8 ± 3.0	77.7 ± 1.8¶¶¶###§&&	79.6 ± 2.1###¶¶¶	81.7 ± 3.5*‡‡¶	83.9 ± 2.7*‡##¶	88.7 ± 2.4***‡‡‡	90.0 ± 1.8***‡‡‡

Symbols as in Table 1.

(878 ± 22 vs. 937 ± 18 ms, $p < 0.05$) and an increase in the power of LF both in absolute and relative terms (6.24 ± 0.39 vs. 5.58 ± 0.41 ln − ms^2, $p < 0.05$ and 57.0 ± 5.7 vs. 41.0 ± 8.6 nu, $p < 0.05$).

Respiration. Compared with spontaneous breathing, reading silently significantly increased the mean respiratory frequency (Fig. 1); conversely, the main respiratory frequency decreased significantly and substantially during both conditions associated with speech (reading aloud and free talking) to values that fell into the LF band of RR (around 0.07 Hz, Fig. 1). This determined a large shift of relative power to the HF band of the respiratory spectrum during reading silently and a large shift of power into the LF band of the respiratory spectrum during conditions associated with speech, both in absolute and relative terms (Fig. 1 and 3). Compared with spontaneous breathing, reading silently increased minute ventilation, whereas reading aloud decreased minute ventilation (Fig. 1).

Blood pressure. Compared with spontaneous breathing, both systolic and diastolic BP increased during all these mental activities (reading aloud or silently or free talking) although the extent of the increase was only moderate (and did not reach significance for the diastolic BP) during reading silently (Table 2).

Overall, these results indicate that these three conditions (free talking, reading aloud and silent reading) determined a minor, though clear, increase in sympathetic activity, as evidenced by changes in RR and BP. The change in the respiratory rate caused by verbalization also had a markedly different effect on global RR variability during the same type of task so that there was a large decrease in breathing frequency, which was displaced into the LF band. This clearly had the effect of artificially increasing the LF components of the RR spectrum, whether or not there was other evidence of sympathetic activation. When speech slowed the breathing rate (into the LF band), the crude RR variability (standard deviation) increased, whereas during silent reading the respiratory rate increased, and the RR variability decreased.

Effect of mental arithmetic, silent versus aloud, versus spontaneous breathing and versus reading silently versus reading aloud. *RR interval.* Compared with spontaneous breathing or reading, arithmetic stress markedly decreased RR and the relative proportion of HF, while it increased the relative proportion of LF. The power of HF also decreased in relative terms with respect to controlled breathing (mild mental stress) and also in absolute terms compared with spontaneous breathing. As with reading aloud and free talking, the RR variability increased compared with spontaneous breathing when speech was present (and breathing was slower, see below), whereas it tended to decrease when speech was absent (and breathing rate was faster).

Respiration. During both conditions of arithmetic mental stress, silent or aloud, minute ventilation appeared slightly, but not significantly, reduced compared with spontaneous

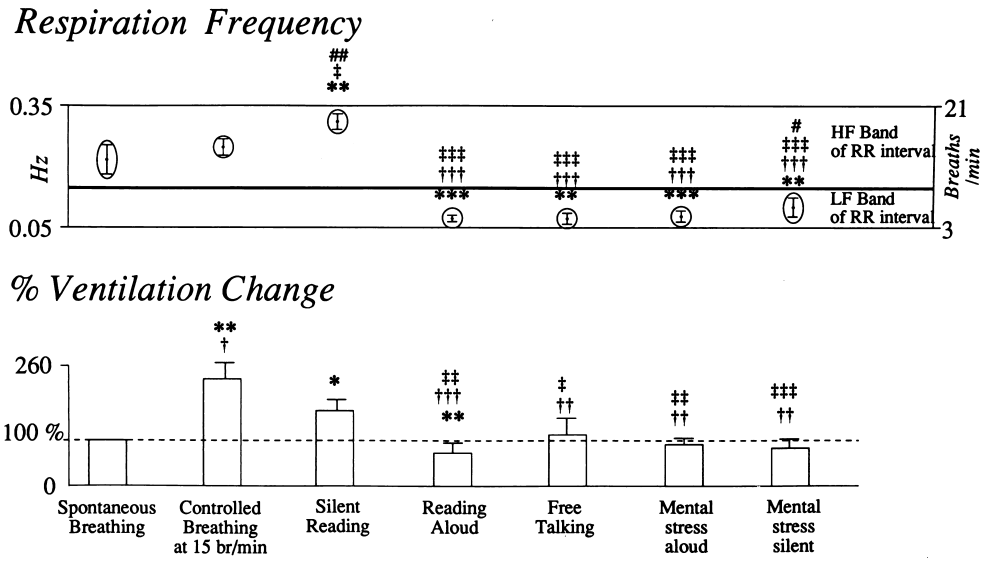

Figure 1. Effects of spontaneous and controlled breathing, reading (mentally or aloud), talking and mental stress (with or without verbalization) on respiratory frequency and on relative changes in minute ventilation (100% corresponds to minute ventilation during spontaneous breathing). Note that, in conditions associated with verbalization or mental stress, respiration is slowed down and falls into the LF band of RR interval (0.03–0.15 Hz). HF = high frequency; LF = low frequency. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$ versus spontaneous breathing; †$p < 0.05$; ††$p < 0.01$; †††$p < 0.001$ versus silent reading; ‡$p < 0.05$; ‡‡$p < 0.01$; ‡‡‡$p < 0.001$ versus controlled breathing; #$p < 0.05$; ##$p < 0.01$ versus mental stress aloud.

breathing (Fig. 1). The frequency of breathing decreased during mental stress with respect to spontaneous breathing, and fell into the LF band. This had the effect of increasing the LF in the respiratory spectrum in both absolute and relative terms and decreasing the HF in relative terms (Fig. 1 and 3).

Blood pressure. Both systolic and diastolic BP increased significantly with arithmetic mental stress, silent or aloud.

Overall, these results indicated a greater degree of sympathetic activation associated with the two types of arithmetic mental stress. It is of interest that the results obtained were rather similar to each other, regardless of speech, due

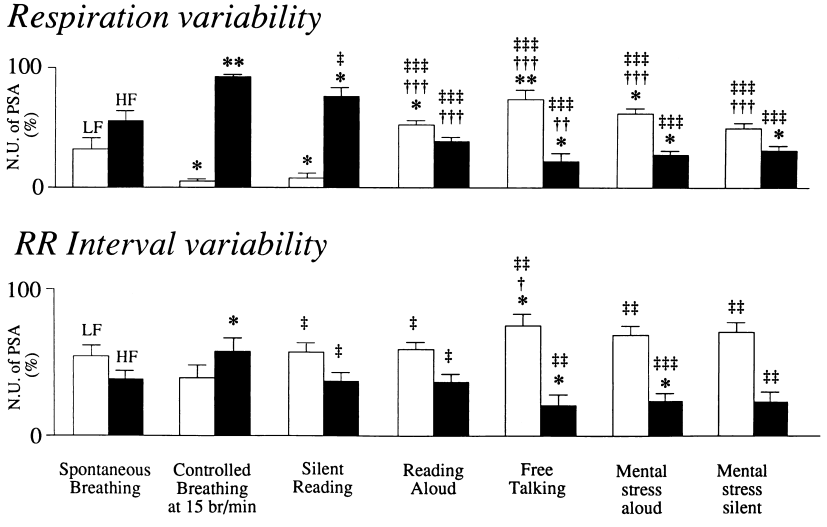

Figure 2. Results (in normalized units, n.u.) of power spectrum analysis (PSA) of respiration and RR intervals (RR) during spontaneous and controlled breathing, reading (mentally or aloud), talking and mental stress (with or without verbalization). Note the inversion of the LF/HF ratio both in RR and respiration, mainly due to the increase in LF **(white bars)** as a consequence of slowing of breathing frequency or sympathetic activation (during mental stress), during reading aloud, free talking and mental stress, compared with spontaneous breathing. LF = low frequency, **open bars;** HF = high frequency, **solid bars.** *$p < 0.05$; **$p < 0.01$ versus spontaneous breathing; †$p < 0.05$; ††$p < 0.01$; †††$p < 0.001$ versus silent reading; ‡$p < 0.05$; ‡‡$p < 0.01$; ‡‡‡$p < 0.001$ versus controlled breathing.

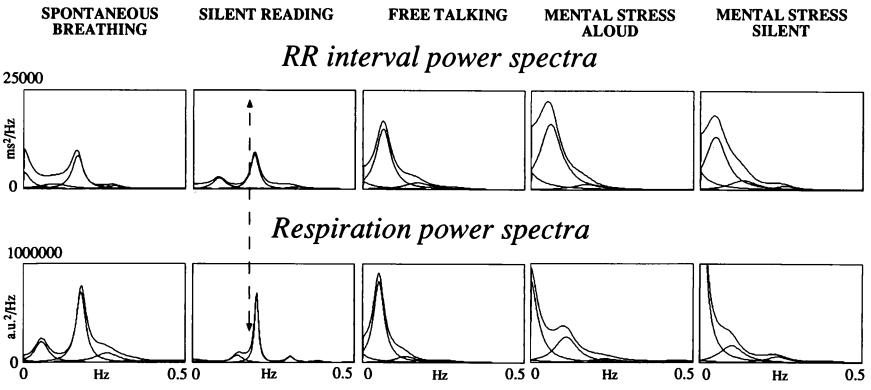

Figure 3. Power spectra of RR interval (RR) and respiration obtained in one subject during spontaneous breathing, free talking and mental stress aloud and silently. During free talking and mental stress aloud, the frequency of breathing decreases, but during mental stress aloud this fact cannot be totally responsible for the increase in the low frequency components in RR because of the evident decrease in respiratory power (i.e., decrease in tidal volume) and the lack of exact coincidence between the low frequency peaks in respiration and RR. Conversely, the increase in the low frequency components in RR during free talking can be explained, to a great extent, by the slowing of respiratory frequency (no decrease in respiratory power and tidal volume). a.u. = arbitrary units; ms = milliseconds.

to a similar increase in sympathetic arousal, also with a shift of the respiratory rate into the LF band of RR.

DISCUSSION

This study clearly shows that during activities such as reading or mental arithmetic, either aloud or silently, the respiratory changes produced by speech markedly alter the variability and the spectral content of the RR, as a result of the change in respiratory pattern necessarily imposed by speech.

Controlled versus free breathing. The change from spontaneous to controlled breathing at rest produces little hemodynamic change. Despite the expected marked increase in HF in the controlled-breathing respiratory spectrum, there is only a modest increase in HF in the RR spectrum. This discrepancy together with the slight decrease in RR might infer less increase in vagal tone or even a modest sympathetic activation, possibly due, in our subjects, to the task of correctly controlling the breathing.

The effect of silent reading on HRV and sympathovagal balance. Compared with spontaneous breathing, reading silently increased respiratory rate and increased ventilation without changes in respiratory power, hence, with little or no change in tidal volume. RR decreased significantly during silent reading, together with an increase in RR-LF and a decrease in HF (nu). So despite similar dominance of respiratory HF, the RR spectrum in silent reading showed more LF than during controlled (or spontaneous) breathing, suggesting a higher degree of sympathetic activation. Due to the higher speed of breathing, silent reading did not cause respiratory interference in the LF band of the RR.

Comparison of reading aloud versus silent reading and free talking. Reading aloud caused a slowing in respiratory rate and a relative reduction in ventilation, with a concomitant increase in respiratory LF. However, the RR spectrum changed very little with respect to silent reading, still showing dominance of LF.

It, thus, appears that the respiratory component of RR did not parallel the increased HF of respiration, most likely due to the effect of the increased sympathetic activity, which increased the LF under both conditions of reading.

Free talking had similar effects to reading aloud, namely a slowing in respiratory rate and a relative reduction in ventilation (12), with a concomitant increase in respiratory LF; RR was also decreased. Nevertheless, as in other conditions associated with slow breathing evaluated in this study, the RR variability was not reduced and actually showed a trend toward an increase (though without reaching statistical significance). This again confirmed previous observations (4) that RR variability is not simply a marker of vagal activity but depends on a complex interaction between vagal and sympathetic activity and respiration.

These results also show that both respiration and sympathetic activity are capable of profoundly affecting the LF (and the HF) components of RR variability. When sympathetic activity predominates, as in conditions associated with high degrees of stress, the LF of RR predominate regardless of the changes in respiration. In the absence of stress, respiration alone is capable of completely altering the RR spectrum by increasing the LF component, i.e., during slow breathing (5,6). In conditions such as reading aloud or free talking, a variable degree of sympathetic activation often interacts with the concomitant effect of slowing of breathing, creating a predominance in the LF component, which is probably partly explained by the increased sympathetic activity and partly by the slowed respiration.

Mental arithmetic, silent versus aloud. As expected, the stress of hurried mental arithmetic produced the greatest dominance of LF in the RR spectrum, compared with the

other activities; this could also be inferred by the increase in BP and by the decrease in mean RR. Figure 2 shows, at first somewhat surprisingly, that free talking produced the highest percentage of LF. This may be explained by the effect of the slower respiratory patterns on HRV. In fact, although there is no doubt that mental arithmetic was more stressful than simple free talking, both free talking and mental arithmetic aloud reduced breathing rates to frequencies in the range of LF. As such the RR spectrum showed in both cases a predominance in LF. The RR decreased much more during mental arithmetic than with free talking, clearly marking the different levels of sympathetic activation between the two conditions.

Thus, it happens that the balance between LF and HF is a composite of sympathetic nervous increase with stress (particularly during silent stress) modified by additional LF added by slower breathing rates (as in free talking). This pattern is similar to that seen during reading aloud versus silent reading.

Study limitations. In this study the need to quantify respiration conflicted with the other need to obtain spontaneous conditions during the various conditions tested and, particularly, with the need to study the effects of speech. Therefore, we could not use a facial mask to quantify ventilation and respiratory gases precisely but had to rely upon an indirect measure of respiration. Inductive belt plethysmography bears a good relationship to changes in lung volume and could be precalibrated against a pneumotachograph. Nevertheless, particularly during talking, the estimates of ventilation are to be considered as only approximate. The results of this study should be applied cautiously to cardiac patients, who are older than the patients studied here, have diseased hearts and possibly concomitant diseases and are on medication. It might be that the effects of the interventions used in this study may differ somewhat between normal subjects and patients.

Conclusions. In conclusion we have found a complex interplay between mental activities with and without speech and various degrees of stress, and the respiratory pattern. These factors in turn alter HRV in two directions: a) mental activity appears invariably associated with some degree of sympathetic activation, whose degree likely depends upon the amount of stress involved in performing the mental task. The purpose of this study was not to measure the level of stress, so we limited our recordings to simple but different conditions: controlled breathing, reading a "neutral" text or performing mental arithmetic. In agreement with previous reports (13–16) we have found that these maneuvers caused different levels of sympathetic activation, as could be seen by the increase in heart rate (decrease in RR), increase in BP and decrease in HRV (standard deviation of RR). This, however, was clearly evident only for those conditions in which interference with the breathing pattern could be excluded. b) The slowing of breathing observed with mental stress had the invariable effect of generating an increase in LF components in the RR power spectrum, regardless of the amount of stress involved in the mental task performed. This was clearly evident by the similarity in RR spectra seen during simple free talking, reading aloud and mental arithmetic, all showing a marked predominance in LF, despite obvious differences in mean heart rate. The slowing of breathing per se thus generated a confounding effect on the RR spectrum, by bringing respiratory sinus arrhythmia (a predominantly vagal effect) into the nonrespiratory LF, thought (simplistically) to reflect sympathetic activity.

These results have practical relevance in the analysis and interpretation of Holter electrocardiograms using HRV techniques. Respiration is not normally measured or analyzed, nor is account taken of such minimal activities (as talking, reading or mental tasks), which we show can perturb the RR spectra to a large extent. One should, therefore, be cautious in the interpretation of short sequences of data (in the range of 4 to 10 min) and not uncritically attribute an increase in LF to sympathetic activation, as this increase might simply be the effect of speech-induced slower breathing. Alternatively, the normal sympathetic activation seen during mental tasks might be masked or accentuated by addition of frequencies generated by unequal or slow respiratory patterns. Thus, in the absence of simultaneous analysis of respiration, the changes in LF/HF ratio should not be taken as clear evidence of changes in autonomic tone.

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REFERENCES

1. Nolan J, Batin PD, Andrews R, et al. A prospective study of heart rate variability and mortality in chronic heart failure: results of the UK Heart Failure Evaluation and Assessment of Risk Trial (UK-HEART). Circulation 1998;98:1510–6.
2. LaRovere MT, Bigger JT, Jr, Marcus FI, et al. Baroreflex sensitivity and heart rate variability is prediction of total cardiac mortality after myocardial infarction. Lancet 1998;351:478–84.
3. Malik M. Task force of European Society of Cardiology and the North American Society of Pacing and Electrophysiology: heart rate variability, standards of measurement, physiological interpretation and clinical use. Circulation 1996;93:1043–65.
4. Bernardi L, Valle F, Coco M, et al. Physical activity influences heart rate variability and very low frequency components in Holter electrocardiograms. Cardiovasc Res 1996;32:234–7.
5. Bernardi L, Keller F, Sanders M, et al. Respiratory sinus arrhythmia in the denervated human heart. J Appl Physiol 1989;67:1447–55.
6. Sanderson JE, Yeung DTK, Yeung LYC, et al. Impact of changes in respiratory frequency and posture on power spectral analysis of heart rate and systolic blood pressure variability in patients with heart failure and normals. Clin Sci 1996;91:35–43.
7. Conway J, Boon N, Jones JV, Sleight P. Involvement of the baroreceptor reflexes in the changes in blood pressure with sleep and mental arousal. Hypertension 1983;5:746–8.
8. Bernardi L, Bianchini B, Spadacini G, et al. Demonstrable cardiac reinnervation after human heart transplantation by carotid baroreflex modulation of RR. Circulation 1995;92:2895–903.
9. Bernardi L, Spadacini G, Bellwon J, et al. Influence of breathing frequency and pattern on oxygen saturation and exercise performance. Lancet 1998;351:1308–11.

10. Piepoli M, Sleight P, Leuzzi S, et al. Origin of respiratory sinus arrhythmia in conscious humans: an important role for the arterial carotid baroreceptors. Circulation 1997;95:1813–21.
11. Malliani A, Pagani M, Lombardi F, Cerutti S. Cardiovascular neural regulation explored in the frequency domain. Circulation 1991;84: 482–92.
12. Winkworth AL, Adams RD, Ellis E. Breathing pattern during spontaneous speech. J Speech Hear Res 1995;38:124–44.
13. Langewitz W, Ruddel H. Spectral analysis of heart rate variability under mental stress. J Hypertens 1989;7 Suppl 6:32–3.
14. Pagani M, Mazzuero G, Ferrari A, et al. Sympathovagal interaction during mental stress. A study using spectral analysis of heart rate variability in healthy control subjects and patients with a prior myocardial infarction. Circulation 1991;83 Suppl 2:43–51.
15. Pagani M, Furlan R, Pizzinelli P, et al. Assessment of the neural control of the circulation during psychological stress. J Aut Nerv Sys 1991;35:33–42.
16. Signorini MG, Saliani V, Lucini D, et al. Parameters from deterministic chaos in the processing of respiration signals during mental stress. J Ambul Monitoring 1992;5:139–52.