Time delay of vagally mediated cardiac baroreflex response varies with autonomic cardiovascular control

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Keyl, C., A. Schneider, M. Dambacher, and L. Bernardi. Time delay of vagally mediated cardiac baroreflex response varies with autonomic cardiovascular control. J Appl Physiol 91: 283–289, 2001.—To examine whether changes in autonomic activity have an effect on the latency of the vagally mediated cardiac baroreflex response in humans, we investigated the effects of neck suction fluctuating sinusoidally at 0.2 Hz on R-R intervals (known to be mediated mainly by vagal activity) in the supine position, during 15° head-down tilt and 60° head-up tilt, and during vagotonic (2 µg/kg) and vagolytic (10 µg/kg) doses of atropine while the subjects breathed at 0.25 Hz. The phase shift between fluctuations in neck chamber pressure and in R-R interval was calculated by complex transfer function analysis and was used as a measure of the time delay between carotid baroreceptor stimulation and cardiac effector response. Cardiac baroreflex responsiveness increased significantly during low-dose atropine and decreased during head-up tilt or 10 µg/kg atropine. With increasing tilt angle, the time delay between cyclic baroreceptor stimulation and oscillations in R-R interval increased from 0.32 ± 0.27 s (head down), to 0.59 ± 0.25 s (supine position, P < 0.05 vs. head down), and to 0.86 ± 0.27 s (head up, P < 0.01 vs. supine). Low-dose atropine had a similar effect to head-down tilt on baroreflex latency, whereas 10 µg/kg atropine increased the time delay markedly to 1.24 ± 0.30 s. Our results demonstrate that changes in autonomic activity, generated either by gravitational stimulus or by atropine, not only affect baroreflex responsiveness but also have a major influence on the latency of the vagally mediated carotid baroreceptor-heart rate reflex. The prolonged baroreflex latency during decreased parasympathetic function may contribute to an unstable regulation of heart rate in patients with cardiac disease.

autonomic nervous system; heart rate; neck suction; spectral analysis

THE INVESTIGATION OF AUTONOMIC cardiovascular regulation has received increasing interest for several reasons, not the least of which is the fact that disturbed autonomic cardiocirculatory control may be associated with increased mortality in patients with heart disease (7, 20). Recently the question arose whether different levels of autonomic activity may affect the time delay of the cardiac baroreflex response (10). This aspect may have relevant implications, because not only the magnitude, but also the time delay, of an effector response may contribute to the stability of a physiological control system, which may be affected by changes in autonomic activity (9, 19, 21, 31). Consequently, a loss of stability in cardiovascular control may be a causative factor in the origin of acute cardiovascular events (35). The latency of the human baroreflex response has been investigated by several authors using different methods (8, 12, 13, 28, 34), but a modulation of the time delay of the vagally mediated cardiac baroreflex response has not been reported before. In the present study, we therefore tested the hypothesis that changes in vagal activity may influence not only cardiac baroreflex sensitivity but also the temporal relationship between baroreceptor stimulation and heart rate response. We stimulated the carotid baroreceptors by sinusoidal cyclic neck suction at a frequency at which heart rate is known to be nearly entirely controlled by vagal activity (1, 5, 31). We performed measurements during different levels of autonomic activity that were generated by a natural stimulus, i.e., changes in body position, and by the application of vagotonic and vagolytic doses of atropine. The effects of baroreceptor stimulation on heart rate were assessed by spectral analytical methods. The phase shift between fluctuations in neck chamber pressure and in R-R interval was used as a measure of the time delay between stimulation of the carotid baroreceptors and the cardiac effector response.

METHODS

After approval of the local Ethical Board, 14 healthy subjects (12 men and 2 women, aged 30–38 yr) were enrolled into the study with their written informed consent. The subjects were studied in the afternoon after training the breathing pattern. We recorded electrocardiogram (Sirecust 302D, Siemens, Erlangen, Germany), noninvasive blood pressure (photoplethysmograph Finapres, Ohmeda, Louisville, CO), respiration (inductance plethysmograph), and neck chamber pressure. Neck suction was performed using a lead collar by

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which a pressure was applied that changed sinusoidally between 0 and \(-30\) mmHg at 0.2 Hz. The subjects were studied in the supine position, at 15° head-down tilt, and at 60° head-up tilt. After a minimum of 10 min for accommodation, 3-min recordings with and without neck suction were performed while the subjects breathed at a fixed frequency of 0.25 Hz. Additionally, seven subjects (6 men and 1 woman, aged 32–42 yr) were studied in the supine position using the identical protocol of neck suction under control conditions, 10 min after application of 2 \(\mu\)g/kg atropine (which produces a vagotonic effect), and 10 min after injection of an additional bolus up to a total dosage of 10 \(\mu\)g/kg atropine (which causes a vagolytic action).

Signals were digitized by a 12 bit analog-to-digital converter and sampled at 1,000 Hz on a personal computer. R waves, systolic blood pressure, and diastolic blood pressure were automatically detected and visually inspected. Analysis of data was performed in accordance with the suggestions of the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (38) and is comparable to the methodology used by other groups (10, 30, 39). Time series were computed with the pressure of neck suction and the respiratory signal both being registered at the start of the R-R interval. Stationarity of each period was checked by the reverse arrangement test (2). Data were resampled at 4 Hz using a moving 500-ms-wide rectangular window (4). After subtraction of the mean value of the sample data, removal of residual linear trends, and application of a cosine tapering window, discrete Fourier analysis was performed for three 50% overlapping windows and the results subsequently averaged. The area under the curve was calculated for the frequency components between 0.175 and 0.225 Hz (frequency component of baroreceptor stimulation) and between 0.225 and 0.275 Hz (frequency component of respiration). The accuracy of the linear relationship between two signals was measured by the coherence function, which was computed using six 50% overlapping windows. A squared coherence >0.53 was accepted as significantly different from zero (39) and interpreted as a sign of stable phase shift. The relationship between the parameters of interest was evaluated by computing the complex transfer function, including gain and phase factors, using six 50% overlapping blocks. According to Bendat and Piersol (3), we estimated the accuracy (i.e., the random error \(e\)) of the estimation of the phase factor \(\hat{\phi}_{xy}(f)\) by

\[
e[\hat{\phi}_{xy}(f)] \approx \frac{1 - \gamma_{xy}(f)^2}{\gamma_{xy}(f)} \sqrt{2n_d}
\]

where \(f\) is frequency, \(\gamma_{xy}(f)\) is the coherence, \(\gamma_{xy}^2(f)\) is the squared coherence, and \(n_d\) is the number of the subrecord blocks. This expression equals the normalized random error of the gain estimate (3). Statistical analysis was performed using commercially available software (SPSS for Windows 9.0; SPSS Chicago, IL). The results of spectral power analysis were normally distributed after logarithmic transformation. Data are presented as means \(\pm\) SD and were compared using a general linear model procedure (repeated-measures analysis of variance, the within-subjects factors were tested by repeated contrasts). The relationship between tilt angle and baroreflex latency was evaluated using linear regression analysis. An \(\alpha\)-error of <0.05 was considered significant.

**RESULTS**

An exemplary registration of R-R intervals during simultaneous breathing at 0.25 Hz and neck suction at 0.25 Hz is demonstrated in Fig. 1. Table 1 summarizes the effects of the tilt maneuvers, and Table 2 reports the effects produced by atropine.

Mean R-R interval decreased progressively from head-down to head-up tilt with significant differences between postures. Mean values of systolic and diastolic blood pressure did not change significantly during changes in body position or application of neck suction.

Neck suction caused an increase in mean R-R interval; however, the relative changes of mean R-R interval between the different body positions were similar, regardless of whether or not simultaneous neck suction was performed. Spectral power of R-R intervals around 0.25 Hz, which may serve as a measure of respiratory sinus arrhythmia, decreased significantly during head-up tilt. This respiratory component of R-R interval spectral power increased during simultaneous neck suction at 0.2 Hz (test of the within-subjects factor neck suction, \(P < 0.05\)), despite the power of the respiratory signal at 0.25 Hz remaining unchanged (Table 1). Comparable to the spectral power of R-R intervals around 0.25 Hz, spectral power around 0.2 Hz decreased significantly during upright position.

Low-dose (2 \(\mu\)g/kg) atropine created a significant increase in R-R interval length, whereas blood pressure remained stable. Higher dose (10 \(\mu\)g/kg) atropine generated a significant decrease in R-R intervals and a slight increase in blood pressure. During the control condition, and during low-dose atropine, neck suction caused a significant increase in the mean R-R interval, as well as in the respiratory component of the R-R interval spectrum, whereas neck suction had no effect on the mean R-R interval and on spectral power around 0.25 Hz during high-dose atropine. The respiratory component of the R-R interval spectrum increased significantly during low-dose atropine and decreased markedly during high-dose atropine. The effects of neck suction on the oscillations of R-R intervals were influenced by atropine in a similar manner: the component around 0.2 Hz increased significantly during low-dose atropine, whereas a marked reduction in spectral power around 0.2 Hz could be observed during high-dose atropine.

Coherence analysis revealed a statistically significant relationship between neck chamber pressure and fluctuation of R-R intervals at 0.2 Hz in all subjects. The squared coherence between neck chamber pressure and R-R intervals was 0.91 \(\pm\) 0.08 during head-down tilt, 0.94 \(\pm\) 0.05 in the supine position, and 0.93 \(\pm\) 0.07 during head-up tilt. The measurements under atropine revealed a squared coherence of 0.95 \(\pm\) 0.04 during the control condition, 0.93 \(\pm\) 0.06 during low-dose atropine, and 0.95 \(\pm\) 0.04 during high-dose atropine. Considering the mean values of the squared coherence, the average random error of the phase factor estimation was 0.095 rad during head-down tilt, 0.075 rad in supine position, 0.082 rad during head-up tilt, 0.088 rad during 2 \(\mu\)g/kg atropine, 0.075 rad during the condition without atropine, and 0.075 rad during 10 \(\mu\)g/kg atropine.
The phase relationship between neck chamber pressure and R-R intervals was 2.74 ± 0.33 rad during head-down tilt, 2.40 ± 0.32 rad during supine position, and 2.06 ± 0.33 rad during head-up tilt. Because a decrease in neck chamber pressure generates an increase in transmural carotid pressure, the phase relationship between changes in neck chamber pressure and changes in R-R intervals is correctly expressed after subtraction of half a period, i.e., after subtraction of \( \pi \). The phase relationship obtained thus was used to calculate the temporal relationship (time delay) between neck suction and fluctuations in R-R interval. With increasing tilt angle, the latency between cyclic baroreceptor stimulation and oscillations in the R-R interval increased from 0.32 ± 0.27 s (head down), to 0.59 ± 0.25 s (supine position, \( P < 0.05 \) vs. head down), and to 0.86 ± 0.27 s (head-up, \( P < 0.01 \) vs. supine position) (Fig. 2). Linear regression analysis demon-

Table 1. Hemodynamic data and logarithmically transformed spectral power of respiration and of R-R intervals at the frequency of respiration (0.25 Hz) and of neck suction (0.2 Hz)

<table>
<thead>
<tr>
<th></th>
<th>15° Head Down</th>
<th>Supine</th>
<th>60° Head Up</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Neck suction</td>
<td>Control</td>
</tr>
<tr>
<td>Mean R-R interval, ms</td>
<td>982 ± 134</td>
<td>1047 ± 132‡</td>
<td>913 ± 128*, 990 ± 166*‡</td>
</tr>
<tr>
<td>Mean SBP, mmHg</td>
<td>101 ± 10.3</td>
<td>103 ± 7.8</td>
<td>109 ± 15.8, 98 ± 17.9</td>
</tr>
<tr>
<td>Mean DBP, mmHg</td>
<td>51 ± 4.9</td>
<td>55 ± 6.3</td>
<td>59 ± 9.9, 54 ± 10.2</td>
</tr>
<tr>
<td>Respiration, ln AU²</td>
<td>−5.72 ± 0.88</td>
<td>−5.59 ± 0.85</td>
<td>−5.19 ± 0.74, −5.37 ± 1.00</td>
</tr>
<tr>
<td>R-R interval spectral power (0.225 to &lt;0.275 Hz), ln (ms²)</td>
<td>5.51 ± 1.26</td>
<td>5.95 ± 1.59‡</td>
<td>5.20 ± 1.83, 5.83 ± 1.61‡</td>
</tr>
<tr>
<td>R-R interval spectral power (0.175 to &lt;0.225 Hz), ln (ms²)</td>
<td>6.86 ± 2.04</td>
<td>6.50 ± 1.76</td>
<td>5.51 ± 1.51†</td>
</tr>
</tbody>
</table>

Values are means ± SD. SBP, systolic blood pressure; DBP, diastolic blood pressure; AU, arbitrary units. *P < 0.05 vs. head down. †P < 0.05 vs. supine. ‡P < 0.05 vs. condition without neck suction (test of the within-subjects factor “neck suction”).
strated a significant relationship between tilt angle and latency in the cardiac baroreflex response ($r = 0.62, P < 0.05$). The analysis of the atropine group revealed a phase angle between neck suction and R-R interval fluctuations at 0.2 Hz of 2.29 ± 0.48 rad during the control condition, 2.65 ± 0.39 rad during 2 μg/kg atropine, and 1.58 ± 0.38 rad during 10 μg/kg atropine. On the basis of these data, the time delay between baroreceptor stimulation and oscillations of R-R intervals increased with decreasing parasympathetic activity from 0.39 ± 0.31 s (2 μg/kg atropine), to 0.67 ± 0.38 s (control, $P < 0.01$ vs. low-dose atropine), and to 1.24 ± 0.30 s (10 μg/kg atropine, $P < 0.01$ vs. control) (Fig. 3).

**DISCUSSION**

Our study demonstrates that changes in the vagally mediated cardiac baroreflex responsiveness, induced by postural changes, as well as by the vagotonic and vagolytic effects of atropine, are associated with changes in the time delay between baroreceptor stimulation and heart rate response.

**Baroreflex sensitivity.** Cardiac baroreflex responsiveness decreased significantly during head-up tilt, as well as during high-dose atropine. Head-down tilt caused only a slight increase, whereas 2 μg/kg atropine generated a much more marked increase in baroreflex responsiveness. These results are in accordance with previous studies that investigated the effects of posture on baroreflex responsiveness and found that cardiac baroreflex sensitivity decreased during upright position (10, 17, 29, 36), whereas cardiac baroreflex sensitivity increased during head-down tilt (17). Other authors did not find changes in baroreflex sensitivity during postural changes (15, 16, 18). Differences in the study designs and methodological differences in the assessment of the cardiac baroreflex response may have contributed to these contrasting results. Furthermore, changes in sympathetic activity have been shown to modify the vagally mediated baroreflex sensitivity during postural changes (14–16).

It is well known that the cardiac autonomic response depends on the frequency of the input signal (6, 26, 31). Multiple studies have demonstrated that fluctuations in heart rate above 0.15 Hz are mainly mediated by
vagal activity (1, 27, 31). Similarly, stimulation of the carotid baroreceptors at frequencies above 0.15 Hz acts mainly by modulation of vagal activity (6, 11, 37). In accordance with this phenomenon, we observed that increases or decreases in parasympathetic activity were accompanied by increases or decreases in R-R interval fluctuations at a respiratory frequency of 0.25 Hz as well as at a frequency of neck suction frequency of 0.2 Hz. Thus our results demonstrate changes in the sensitivity of the vagally mediated cardiac baroreflex response.

**Time delay of the effector response.** Previous studies reported a baroreflex latency of 500–600 ms in humans (8, 28), which is similar to our results obtained in the supine position, whereas other authors observed significantly shorter latencies (12). Besides the fact that we did not measure the P-wave response but did measure changes in R-R intervals, our method differed from these studies that used either pharmacological blood pressure changes, electrical nerve stimulation, or abrupt neck suction stimuli. Unlike abrupt baroreceptor activation by means of brief stimuli, which is directly dependent on the modulation of baroreflex sensitivity by the respiratory and cardiac phase (13, 34), cyclic stimulation of carotid baroreceptors may provide an average estimation of the cardiac baroreflex responsiveness. The use of frequency-domain methods allows the assessment of the phase shift between input (oscillation in neck chamber pressure at 0.2 Hz) and output (oscillation in R-R interval at 0.2 Hz) independently of oscillations in the R-R interval at other frequencies and, which may be much more important, independently of the gain of the baroreflex. Following the above-mentioned frequency dependency of autonomic cardiac control, our approach demonstrates that the latency of the vagally mediated cardiac baroreflex response undergoes marked variations in response to changes in vagal activity.

Furthermore, our findings suggest an evident relationship between the tilt angle and the latency of the baroreflex response. Whereas a modulation of baroreflex latency has been clearly demonstrated in muscle sympathetic nerves (40), changes in the vagally mediated baroreflex latency have not been reported until now. Following previous studies, it may be hypothesized that the latency of the vagally mediated cardiac baroreflex response is related to the autonomic motoneuron responsiveness, which varies with different postures (10).

**Effects of atropine.** The results obtained during atropine administration confirm the measurements during head-up and head-down tilt: the vagotonic effect of low-dose atropine created an increase in respiratory sinus arrhythmia, as well as in cardiac baroreflex responsiveness, whereas the vagolytic effect was related to a decrease of these two parameters. These findings are in accordance with previous studies that found a decrease and an increase in the high-frequency component during vagotonic and vagolytic doses of atropine, respectively (23), and a totally abolished R-R interval response to neck suction at 0.2 Hz after administration of 40 μg/kg atropine (5). When the effects of atropine and of head-down and head-up tilt are compared, it was clearly evident that the vagotonic and vagolytic effects of atropine on the mean R-R interval, respiratory sinus arrhythmia, and baroreflex sensitivity were much more marked than the effects of the postural changes on these parameters. The shortening of the latency between baroreceptor stimulation and oscillations in R-R interval is comparable between low-dose atropine and head-down tilt, possibly indicating a maximal shortening of the reflex transmission. However, 10 μg/kg atropine showed a much more pronounced vagolytic action than the natural gravitational stimulus, which was reflected by the more marked increase in baroreflex latency during high-dose atropine compared with head-up tilt.

**Respiratory sinus arrhythmia.** A previous study found that respiration and neck suction influenced heart rate independently of each other (11). However, we observed that neck suction at 0.2 Hz was accompanied by an increase in mean R-R interval and an increase in the respiratory fluctuations of the R-R interval at 0.25 Hz regardless of the body position, despite the respiratory signal being unchanged. This phenomenon did not affect the relative changes of the mean R-R interval during postural changes. It cannot be excluded that the breathing pattern was influenced by neck suction in a manner that did not change the breathing signal but that influenced respiratory sinus arrhythmia. On the other hand, our observation may indicate additive effects of the increase in vagal activity during cyclic neck suction and the decrease in vagal activity during head-up tilt.

**Clinical implications.** Several studies revealed a correlation between impaired autonomic cardiac control, assessed by the analysis of heart rate variability or baroreflex sensitivity, and poor outcome in patients with chronic heart disease (7, 20, 33, 35). Even such prognostic indexes like the absence of shortening of the R-R interval immediately after an abrupt drop in arterial blood pressure (caused by a single ventricular ectopic beat) (32) have been explained by a disturbed baroreflex control of heart rate (24). Because our methodological approach revealed that the time delay of the vagally mediated cardiac control may in fact be prolonged during reduced vagal activity, it might be possible that, apart from an impaired gain of the baroreflex, an increase in the time delay of the vagally mediated reflex transmission contributes to phenomena like those observed by Schmidt et al. (32).

Furthermore, it is well known that the time delay, as well as the gain, of the effector response may have a major impact on the stability of a negative-feedback control system (21). An increase in the time delay of the effector response may generate an unstable state of regulation. This interaction has been demonstrated for the sympathetic mediated control of arterial pressure and heart rate (22) but, until now, not for the vagally mediated heart rate response. However, a computer model revealed that changes in the time delay typical for the vagally mediated cardiac control can be related
to major changes in the dynamic behavior of heart rate (9). Whereas the system showed a stable behavior during low values of the time delay, spontaneous oscillations of varying complexity occurred when the time delay was increased (9). Therefore, it is possible that an increased time delay of the cardiac baroreflex response, associated with an impaired parasympathetic function, may contribute to an unstable regulation of heart rate and thus to cardiac rhythm instabilities in patients with chronic heart disease. This question should be subject to further research.

Limitations of the study. Because we analyzed the relationship between fluctuations in neck chamber pressure and R-R interval oscillations, our results may have been influenced by changes in the atrioventricular (AV) conduction time during the postural changes. The variability of the AV interval and its dependency on the body posture have been studied by Nollo and co-workers (25). The authors found that neither the mean AV interval nor the AV interval variability increased significantly during 60° head-up tilt compared with the supine position. When the heart rate was fixed, the mean AV conduction time increased by 25 ms during head-up tilt, whereas the AV interval variability was reduced (25). With respect to these data, one may assume that, even under the condition of baroreflex stimulation (causing fluctuations in R-R intervals comparable to those of respiratory sinus arrhythmia), the influences of changes in body position on the AV conduction time are markedly less than the changes in baroreflex latency observed in our study.

Furthermore, it must be emphasized that this study measures the net effect of the mainly vagally mediated cardiac baroreflex response and cannot differentiate between the central modulation of autonomic activity [possibly an effect of the vagotonic action of atropine (23)] and the peripheral muscarinic blockade.

Conclusion. In conclusion, our study demonstrates that changes in autonomic activity do not only affect vagally mediated cardiac baroreflex responsiveness but also have a major influence on the latency of the carotid baroreceptor-heart rate reflex. The increased time delay of the cardiac reflex response during decreased vagal activity may contribute to the disturbed cardiocirculatory stability in patients with impaired cardiac function.

REFERENCES


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