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(Citation)
European Journal of Applied Physiology,97(2):240-247

(Issue Date)
2006-06

(Resource Type)
journal article

(Version)
Accepted Manuscript

(URL)
https://hdl.handle.net/20.500.14094/90000083

Ms # EJAP-00282-2005 – R2

Phase-dependent chronotropic response of the heart during running in humans

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ABSTRACT

Heartbeat modulation by muscle contraction during rhythmic exercise involving a small muscle mass is phase-dependent, reflecting the timing of the muscle contraction within the cardiac cycle, but it remains unclear whether such modulation occurs during whole body exercise. To determine whether phase-dependent chronotropic changes in the heart would occur during running, we investigated the relationship between R-R interval (RRI) and the timing of vastus lateralis muscle contractions within the cardiac cycle. Seven healthy subjects were examined during high intensity running where the target heart rate was 160 beats·min^{-1}. The running pitch was made to wax and wane periodically in the neighborhood of the target heart rate to scan the effect of footfall timing within the cardiac cycle on heart period. We found that when muscle contraction occurred early in the cardiac cycle, RRI was reduced from the mean RRI ($P < 0.05$). Conversely, when muscle contraction occurred in the latter half of the cardiac cycle, RRI tended to increase ($P > 0.05$). Thus the curve reflecting this phase-dependent relationship between heart period and the timing of muscle contraction showed a positive slope within the first one-quarter to three-quarters of the cardiac cycle. Our results suggest the existence of a mechanism that provides beat-by-beat regulation of RRI even when it is very short (~ 375 msec), i.e., a cardio-locomotor synchronization develops during running when the frequencies of the two rhythms approach one another.

Keywords. entrainment; synchronization; coupling; phase-response curve; rhythmic exercise;

INTRODUCTION

Cardiac rhythm oscillates by entrainment of multiple pacemaker cells (Capelli 1999; Verheijck et al. 1998) as modulated by physiological rhythm(s). The phase-resetting of pacemaker activity for entrainment has been demonstrated not only through analysis of a mathematical model (Michaels et al. 1984) but also through experimentation with the isolated myocardium (Anumonwo et al. 1991). Notably, synchronization of the cardiac and locomotor rhythms (CLS: cardio-locomotor synchronization) is observed during rhythmic exercise (Kirby et al. 1989; Niizeki et al. 1993; Nomura et al. 2003).

Although CLS is observed in humans performing various forms of rhythmic exercise, its properties seem to vary. Kirby et al. (1989, 1990, 1991, 1992) detected CLS during walking, running, cycling, hopping, skipping, and finger tapping. Equally, Udo et al. (1990) and Niizeki et al. (1993) found CLS during walking and running. Still, the CLS is not necessarily the result of physiological interactions. This is because it is possible for synchronization to occur casually when the frequencies of the two independent rhythms approach one another. Taking this into account, the possibility of an occasional occurrence of accidental CLS during hopping, skipping and tapping was not ruled out by earlier investigators (Kirby et al. 1991, 1992) or its likelihood during walking, running and cycling was not evaluated (Kirby et al. 1989; Niizeki et al. 1993; Udo et al. 1990). Using a surrogate data technique, we have ruled out the possibility of accidental CLS during running but not during cycling (Nomura et al. 2001, 2003). To characterize the relationship between cardiac and locomotor rhythms independently in that experiment, we generated surrogate data by means of random shuffle of the series of gait cycle. In that case, if CLS occurred by chance, the surrogate data would have the same characteristics of CLS as the original. After surrogation, however, the relationship between original cardiac rhythm and surrogate locomotor rhythm during running was not indicative of CLS but during cycling; in other words, the possibility of accidental CLS during running was rejected but not during cycling. In addition, the effect of exercise differences on the properties of CLS were not

quantitatively assessed in the aforementioned studies, though the duration of CLS was longer during running than during cycling (Nomura et al. 2003). We also showed that CLS occurred with specific phase differences during running, but with non-specific phase differences during cycling. Thus, the stability of CLS is affected by the type of exercise being performed.

Analysis of the phase-response curve (PRC) between two rhythms enables us to predict the stability of entrainment between two rhythms (Pavlidis 1973), but there has been no evaluation of a PRC obtained from cardiac and locomotor rhythms during natural rhythmic exercise as running. The chronotropic response of the heart to brief muscle contraction is affected by the timing of the contractions within the respiratory (Nakamura et al. 1997) and cardiac cycles (Niizeki and Miyamoto 1998, 1999), and Niizeki and Miyamoto (1999) found that the heartbeat response to the timing of muscle contractions within the cardiac cycle was phase-dependent during handgrip exercise. There was a shortening of the heartbeat interval when muscle contraction occurred early in the cardiac cycle, but a prolongation when the muscle contraction occurred in the latter half of the cardiac cycle. Also, the PRC constructed by fitting a regression line to the experimental data corresponded to the criterion for entrainment derived from the theoretical analysis of Pavlidis (1973). It remains unclear, however, whether the relationship between cardiac and locomotor rhythms during natural rhythmic exercise as running corresponds to the criterion for entrainment. Given that the possibility of accidental CLS during running can be ruled out (Nomura et al. 2001, 2003), we hypothesize that the relationship between cardiac and locomotor rhythms reflects a phase-dependent chronotropic response of the heart to the timing of lower extremity muscle contraction associated with footfalls within the cardiac cycle during running. To test this hypothesis, we investigated the influence of the cardiac phase on the changes in heartbeat interval elicited by the lower extremity muscular activity associated with footfall during running.

METHODS

Seven healthy men [mean height, 168.0 (range 165-171) cm; body mass, 61.2 (52-69.7) kg; age, 22.3 (21-24) years] with no history of cardiopulmonary disease participated in this study. Each subject signed an informed consent statement after being provided with a verbal explanation of the intent and the procedures of the experiments. This study was approved by the Human Subjects Committee of the department at Kobe University.

Protocols

A 10-min warm-up period was followed by at least 15 min of rest during which the subjects were instrumented. Thereafter, each subject participated in a 20-min session in which he ran on a treadmill (NT-12; Nishikawa, Japan) at a pitch that was controlled by computer generated buzzer signals throughout the session.

For the first 4 min of the measurement period, the treadmill speed was gradually increased until a target heart rate of 160 beats·min^{-1} was achieved, and it was then maintained at that speed throughout the remainder of the session. This target heart rate was selected in order to achieve a high intensity workload. During that period, the treadmill speed ranged from 144 to 230 m·min^{-1} (0% grade).

For the first 4 min of the measurement period, the pace of the buzzer signal was fixed at 160 beats·min^{-1}. Then for the remaining 16 min, the pace of the buzzer signal was periodically oscillated to scan the effect of footfall timing within the cardiac cycle on heart period at several cardiac phases. One cycle of the oscillation was fixed at 8 min, which corresponded to approximately 0.002 Hz, and the oscillation amplitude was fixed at 155 to 165 beats·min^{-1}.

Data collection

The beat-to-beat RR interval (RRI) was measured from a surface electrocardiogram (ECG) using standard bipolar leads (CM_5). To distinguish the R waves of the QRS complex, the ECG signal was amplified and filtered (AB-621G; Nihonkoden, Japan). To avoid movement artifacts, we set the filtering frequency band at 10 to 300 Hz. Because the electromyogram (EMG) of the lower extremity shows muscular discharge mostly at initial contact of the foot with the treadmill surface during running, the rhythm of the muscle contraction was regarded as the locomotor rhythm, and we defined the muscle contraction interval (MCI) as the interval between the onset of one muscle contraction and that of the next muscle contraction. The MCI was measured from a surface EMG from the right vastus lateralis muscle using bipolar leads. Surface electrodes were placed along the longitudinal axis of the muscle, approximately 10 cm superior to the head of the fibula to avoid EMG baseline agitation by motion of the leg; the earth electrode was placed on the upper thigh such that the interelectrode distance was about 30 mm. We defined the breathing interval (BRI) as the interval between the onset of one inspiration and that of the next. The BRI was measured from the chest using an uncalibrated impedance spirogram (AI-601G; Nihonkoden, Japan) (Baker and Geddes 1970).

These input signals were digitized using a 12-bit analog-to-digital converter (DACcard500; National Instruments Corporation) sampling at 1,000 Hz and were processed off-line. A customized computer (IBM PC/AT) program detected the QRS spikes in ECG, the onset of muscle contractions in EMG and the onset of inspirations in the thoracic impedance signal. To detect the onset of a muscle contraction, the EMG signal was converted to an integrated EMG with full-wave rectification and smoothed, after which the onset was defined as the time at which the integrated EMG increased above a preset trigger level.

Data analyses

Each successive QRS spike and the onsets of muscle contraction and inspiration were marked as equivalent to one oscillatory cycle, corresponding to which a 2π increment was added. Within this one oscillatory cycle, the instantaneous phase is

$\phi(t) = 2\pi \dfrac{t - t_k}{t_{k+1} - t_k} + 2\pi k$, $\quad t_k \le t < t_{k+1}$, where $\phi(t)$ is the instantaneous phase at *t*, and t_k is time of the *k*-th marker event. With this definition, the phase is a monotonically increasing piecewise-linear function of time defined on a real line.

A phase synchrogram is helpful when evaluating the phase relationship between two rhythms (Rosenblum et al. 1998). To obtain a phase synchrogram, a generalized relative phase has to be calculated. For example, the generalized relative phase (Ψ_{a-b}) of the "b"oscillator for the onset of the "a" oscillator event is calculated as follows.

$\Psi_{a-b} = \dfrac{1}{2\pi}(\phi_b(t_k) \bmod 2\pi)$, where t_k is the time of the *k*-th occurrence of an "a" oscillator event, and ϕ_b is the instantaneous phase of the "b" oscillator. After removing a small number of abnormal RRIs, MCIs and BRIs, the occurrence of the phase synchronization between each oscillation was monitored using a phase synchrogram. For example, the time trace of Ψ_{RR-MC} was visualized by plotting generalized relative phases vs. time (Fig. 1). As mentioned, our target heart rate was 160 beats·min^{-1}, and the locomotor rhythms, which were controlled at 160 (range ± 5) beats·min^{-1} corresponded to 2.66 (range 2.58 to 2.75) Hz. Because we used EMG from only the right vastus lateralis muscle to estimate locomotor rhythms, the estimated rhythm was approximately $1.33 \pm \zeta$ Hz (ζ is the variability of the locomotor rhythm). As a result, the 2 : 1 phase synchronization observed in the phase synchrogram corresponds to 1 : 1 phase synchronization of the cardiac rhythm to the locomotor rhythm (Kirby et al. 1989, 1992), who evaluated locomotor rhythm using an accelerometer placed at the chest.

[Insert Figure 1 here about]

The data obtained between minutes 8 and 16 of the measurement period were evaluated. This period corresponded to one cycle of the periodical oscillation of the locomotor rhythm. The timing of the muscle contractions with respect to the cardiac phase ($\phi_{MC-RR\,ave}$) was calculated as $\phi_{MC-RR\,ave} = (t_{MC} - t_{R-peak}) / \overline{RRI}$, where t_{MC} was the onset time of muscle contraction, t_{R-peak} was the time of the R wave peak, and $\overline{RRI}$ was the average RRI during the analysis period. To evaluate the relationship between the timing of the muscle contractions and the heart period, we plotted changes in heart period as the amount of change in the RRI (ΔRRI) from $\overline{RRI}$ vs. the timing of the muscle contractions.

Statistics

To evaluate the effect of muscle contraction timing within the cardiac cycle on heart period, the average RRI (i.e., $\phi_{MC-RR\,ave}$) was split into ten subintervals of equal length class (class I, II,... X), and the mean value of ΔRRI was calculated for each class. Then for each subject differences in mean ΔRRI among classes were evaluated using one-way analysis of variance (ANOVA). Two-way ANOVA was used to evaluate the effect of class on the group mean ΔRRI, and for multiple comparisons of the values at class II, III,..., and X with those at class I, paired t-tests were used with application of Holm's correction to keep the total error of the tests below 5%. Values of $P < 0.05$ were considered significant.

RESULTS

Figure 2 shows the transient changes in a representative RRI (A), MCI (B) and BRI (C) time series in one subject. The pitch of the beep signal was set at $160\,\text{beats}\cdot\text{min}^{-1}$ for the first 4 min of the measurement period and was then periodically oscillated (8-min cycles) within 160 ± 5 $\text{beats}\cdot\text{min}^{-1}$ for the remaining 16 min. The oscillation frequency of the MCI corresponded to the frequency of the controlled pace (Fig. 2B). BRI did not show a

regular rhythm at any frequency. Phase synchronization was observed between the cardiac and locomotor rhythms during minutes 2-5 and 12-15 of the measurement periods, whereas no obvious phase synchronization was observed between the cardiac and respiratory rhythms or between the locomotor and respiratory rhythms. The synchronization between the cardiac and locomotor rhythms occurred at specific phases (approximately 0.4 and 0.9 of Ψ_{RR-MC}). The locomotor rhythm became $160\,\text{steps}\cdot\text{min}^{-1}$ at minutes 8, 12, 16 and 20 of the measurement period and then decreased or increased gradually. Ψ_{RR-MC} showed a phase drift (i.e., the cardiac rhythm was faster then the locomotor rhythm) during minutes 8-10 of the measurement periods, with a gradual reduction in the locomotor rhythm (i.e., a gradual increase in MCI). By contrast, during minutes 12-14 of the measurement periods there was a gradual increase in locomotor rhythm (i.e., a gradual decrease in MCI), and Ψ_{RR-MC} showed a horizontal alignment. The characteristics of the phase relationship between the rhythms were similar for all seven subjects, but the durations of the horizontal alignment varied.

[Insert Figure 2 here about]

Figure 3 shows the influence of muscle contraction timing within the cardiac cycle on the variation in RRI from the average RRI for all seven subjects. A negative value of ΔRRI indicates a phase advance in the RRI change; conversely, a positive value indicates a phase delay. The changes in ΔRRI depended on the $\phi_{MC-RR_{ave}}$, and one-way ANOVA showed an effect on RRI of muscle contraction timing within the cardiac cycle in all subjects ($P < 0.05$). For example, muscle contractions occurring early in the cardiac phase at $\phi_{MC-RR_{ave}} \approx 0.3$ produced a relative shortening of the RRI in all seven subjects. If it occurred later in the cardiac phase, however, a relative prolongation of the RRI occurred. The magnitudes of shortening and prolongation of the RRI varied among the subjects.

[Insert Figure 3 here about]

Figure 4 shows the group mean and standard error of the phase effective influence of muscle contraction timing within the cardiac cycle on the change in RRI from the average RRI. The phase-dependent changes in RRI showed that ΔRRI at class III and IV, which corresponding to $0.2 < \phi_{MC-RR\,ave} < 0.4$, were lower ($P < 0.05$) than class I (i.e., $0 < \phi_{MC-RR\,ave} < 0.1$). In addition, the PRC had a positive slope within $0.2 < \phi_{MC-RR\,ave} < 0.6$, which corresponds to a negative slope of the PRC.

[Insert Figure 4 here about]

DISCUSSION

To affirm the hypothesis that there is a relationship between cardiac and locomotor rhythms during running that corresponds to the criterion for entrainment between two rhythms (Pavlidis 1973), we must obtain evidence of an influence of cardiac phase on the change in heartbeat interval elicited by a change in footfall timing within the cardiac cycle. According to the theoretical analysis, stable 1 : 1 entrainment can occur in phase-advance regions, where the slope of the PRC obtained from the relationship between the driven and the drive oscillators is more than -2 and less than 0 (Pavlidis 1973). To evaluate the locomotion rhythm during running, we measured the interval between the contractions of the right vastus lateralis muscle. We selected this muscle because it acts with every footfall of the right lower limb. Consequently, we found that there is a positive chronotropic change in the heart when lower extremity muscular activity occurs early in the cardiac phase, and that there is no change, or a small negative chronotropic change, when lower extremity muscular activity occurs in the latter half of the cardiac phase (Figs. 3 and 4). This phase-dependent relationship between chronotropic changes in the heart and muscle contraction timing within the cardiac cycle corresponds to the criterion for entrainment. We believe the present observations of a phase-dependent relationship to confirm CLS during running (Kirby et al. 1989; Niizeki et al. 1993; Nomura et al. 2003).

During handgrip exercise the heart rate response to muscle contraction timing within the cardiac cycle does correspond to the properties of entrainment as defined by the theoretical analysis (Niizeki and Miyamoto 1999). The results of Niizeki et al. (1999) may explain CLS during running, though their results raise two issues. First, it is unclear whether their results can be extrapolated to high intensity running, such as that examined in the present study, because of the dynamics of the changes in heartbeat interval that occur with differences in exercise intensity. And second, the relationship between the original cardiac rhythm without processing (i.e., application of a filter in the frequency domain) and locomotor rhythms is unclear because they removed the fluctuation in heartbeat interval within a specific frequency range. The respiratory sinus arrhythmia (RSA) contributes substantially to heartbeat fluctuations during low intensity exercise. Nevertheless, Niizeki et al. (1999) removed the low frequency components of the heartbeat fluctuations that were below the RSA by applying a rectangular window filter because the frequency of the heartbeat fluctuations relative to the muscle contractions was higher than its RSA. Their technique might be an excellent tool for extraction of objective signals from noisy ones, but we aimed to find evidence of phase-dependent chronotropic changes in the heart elicited by muscle contraction with no processing in the frequency domain because the relationship between the unprocessed cardiac rhythm and the locomotor rhythm develops into CLS during running.

By examining the relationship between cardiac and locomotor rhythms during running with a target heart rate of $160\,\text{beats}\cdot\text{min}^{-1}$, the present study clarified that the chronotropic changes in the heart to the muscle contraction timing within the cardiac cycle was phase-dependent during whole body exercise at the high intensity. In addition, by analyzing the changes in heartbeat interval without processing in the frequency domain, it was clarified that the heartbeat response where any frequency components of the heartbeat fluctuations were included was phase-dependent to the timing of muscle contraction. To more clearly show the heartbeat response to the muscle contraction timing during handgrip exercise, Niizeki et al. (1999) removed the low frequency components below the RSA in

the frequency domain. Although the contribution of RSA to heartbeat fluctuation is notable, the high frequency components relative to RSA are negatively related to exercise intensity (Billman and Dujardin 1990; Grossman et al. 2004; Yamamoto et al. 1991). That is, it is implied that because RSA depends on exercise intensity, the influence of the muscle contraction timing on the heart period response is hidden in the contribution of RSA during low intensity exercise, whereas it appears clearly during high intensity exercise.

According to the theoretical analysis of Pavlidis (1973), the criterion for entrainment is a necessary condition wherein a nonlinear oscillator is entrained to an external oscillator when the frequency of external oscillator approaches that of the nonlinear oscillator. As our results, it was demonstrated that the relationship between cardiac and locomotor rhythms corresponded to the criterion for entrainment. However, physiological parameters are not considered in the criterion for entrainment. Thus, the physiological mechanism(s) underlying the phase-dependent chronotropic change in the heart related to the timing of lower limb muscle contraction associated with footfalls within the cardiac cycle cannot be determined from the results of the present study. But we speculate two possible mechanisms: a peripheral neural circuit and a non-neural mechanism in the following.

A peripheral neural circuit in which ascending signals from mechanoreceptors within active skeletal muscle may modulate the heartbeat interval. Muscle mechanoreceptors are able to respond instantaneously to generate afferent signals in synchrony with repetitive brief muscle contractions (Legramante et al. 2000). In addition, afferent signals originating from muscle mechanoreceptors inhibit the cardiac vagal component of the baroreceptor reflex and play a role in shortening the heartbeat interval (McWilliam and Yang 1991). Although it is thought that parasympathetic nerve activity is largely withdrawn during high intensity exercise, in dogs significant functional parasympathetic activity to the heart continues even during heavy dynamic exercise (O'Leary et al. 1997). If that is also the case in humans, the remaining cardiac vagal nerve discharge might be modulated by afferent signals derived from muscle mechanoreceptors. On the other hand, Niizeki (2005) found

that synchronization between the rhythms of the cardiac and intramuscular pressure changes can be achieved with mechanical thigh compression with no muscle contraction, which implies that muscle contraction is not necessary to modulate heartbeat interval. Perhaps some skeletal muscle mechanoreceptors are able to transduce changes in intramuscular pressure. In addition, Williamson et al. (1995) found that a shortening of the onset RRI was not detected when exercise onset, which was passively induced cycling combined with electrical stimulation (i.e., without central command), occurred in the last one- and two-third of the cardiac cycle. They suggested that the latency of about 600 msec or more is necessary in a shortening of RRI by muscle-heart reflex. However, in the case of a rhythmical muscle contraction, it is uncertain whether this property of muscle-heart reflex is the same.

The second explanation is a non-neural mechanism, such as an intrinsic property of the heart without a modification of autonomic nerve activity . Heartbeat interval was shortened via mechanoelectric feedback within the myocardium stimulated by stretching of the atrial wall associated with diastolic volumetric load (Kohl et al. 1999). Also consistent with the idea that a non-neural mechanism regulates short-term fluctuation of heartbeat interval during heavy dynamic exercise is the finding that ganglionic blockade reduces but does not abolish RSA (Kohl et al. 1999; Casadei et al. 1996). Moreover, even though in heart transplant patient the heart is not modulated by autonomic nerve, heartbeat interval fluctuation with respiration is evident at rest and during exercise (Bernardi et al. 1990). During running, intrathoracic pressure not only changes with respiratory movement, but there are transient decreases with downward diaphragm movement coincident with footfall (Banzett et al. 1992). Also the muscle contraction acts as pump to increase venous return (Sheriff et al. 1993), which means that blood flow in the femoral vein would increase synchronously with muscle contraction (Laughlin and Schrage 1999; Magder 1995). Thus, transient changes in venous return may affect the volumetric load of the right atrium.

During exercise, descending signals from the limbic system (i.e., central command) drives the circulatory system (Mitchell 1990; Spyer 1989; Turner 1991) by inhibiting the aortic baroreceptor-heart rate reflex at the onset of muscle contraction, so that heart rate increases in parallel with blood pressure (Murata et al. 2004). Williamson et al. (1995) found that the onset RRI shortened when the volitional exercise (i.e., central command and muscle-heart reflex) was initiated in all phase of the cardiac cycle, and when the passively induced exercise combined with electrical stimulation (i.e., without central command) was initiated within the last one-third of the cardiac cycle. As their results, they concluded that heart rate changes elicited by central command can occur fast than responses produced by the muscle-heart reflex. In addition, heartbeat fluctuations are modulated by fictive locomotion caused by electrical stimulation of the mesencephalic locomotor region (Kawahara and Yamauchi 1990; Kawahara et al. 1994). These lead us to speculate that a shortening of RRI might occur by central command when muscle contraction occurred early in the cardiac cycle. It is, however, uncertain whether central command synchronizes with the locomotor rhythm.

There are two limitations to the present study. First, we could not ascertain the causes of the individual differences among the subjects in the magnitudes of the maximum positive chronotropic changes in the heart. We believe that these differences are responsible for the individual differences in CLS duration. This is because when the phase-dependent chronotropic change in the heart is larger, the slope of the PRC is steeper, which is indicative of more stable “entrainment” (Pavlidis 1973). Second, we were unable to validate the effect of respiratory rhythm on the relationship between cardiac and locomotor rhythms. However, respiration seems to have little or no effect on the fluctuations of heartbeat interval because the contribution of the RSA to heartbeat fluctuations is negatively related to exercise intensity (Billman and Dujardin 1990; Grossman et al. 2004; Yamamoto et al. 1991). It should also be added that the other respiratory effect may influence cardiac response. Synchronization between locomotor and respiratory rhythms occurs during rhythmic exercise such as cycling, running and rowing (Bechbache and

Duffin 1977; Bernasconi and Kohl 1993; Bramble and Carrier 1983; Mahler et al. 1991; Paterson et al. 1986). The individual differences among the subjects in the magnitude of chronotropic response of the heart to the timing of muscle contraction might be influenced by the existence of synchronization between locomotor and respiratory rhythms. Further study would be needed to test the respiratory effects.

We have demonstrated positive chronotropic changes in the heart that occur when lower extremity muscular contraction associated with footfall during running occurs early in the cardiac cycle. These findings are consistent with the criterion for entrainment that occurs when frequencies of two rhythms approach one another. Our findings enable us to out the possibility of an accidental CLS during running completely; moreover, they are consistent with the idea that CLS does depend on the type of exercise being performed, and that the variation is explained by the phase-dependent relationship between the cardiac and locomotor rhythms. Still, the mechanism(s) by which different exercises affect fluctuations in heartbeat interval remains unclear.

ACKNOWLEDGEMENTS

We are sincerely grateful to Dr. Matsukawa, K. (Department of Physiology, Graduate School of Health Sciences, Hiroshima University, Hiroshima, Japan), Dr. Ando, H. (Faculty of Health Science, Kobe University School of Medicine, Kobe, Japan) for their useful suggestions and valuable discussions.

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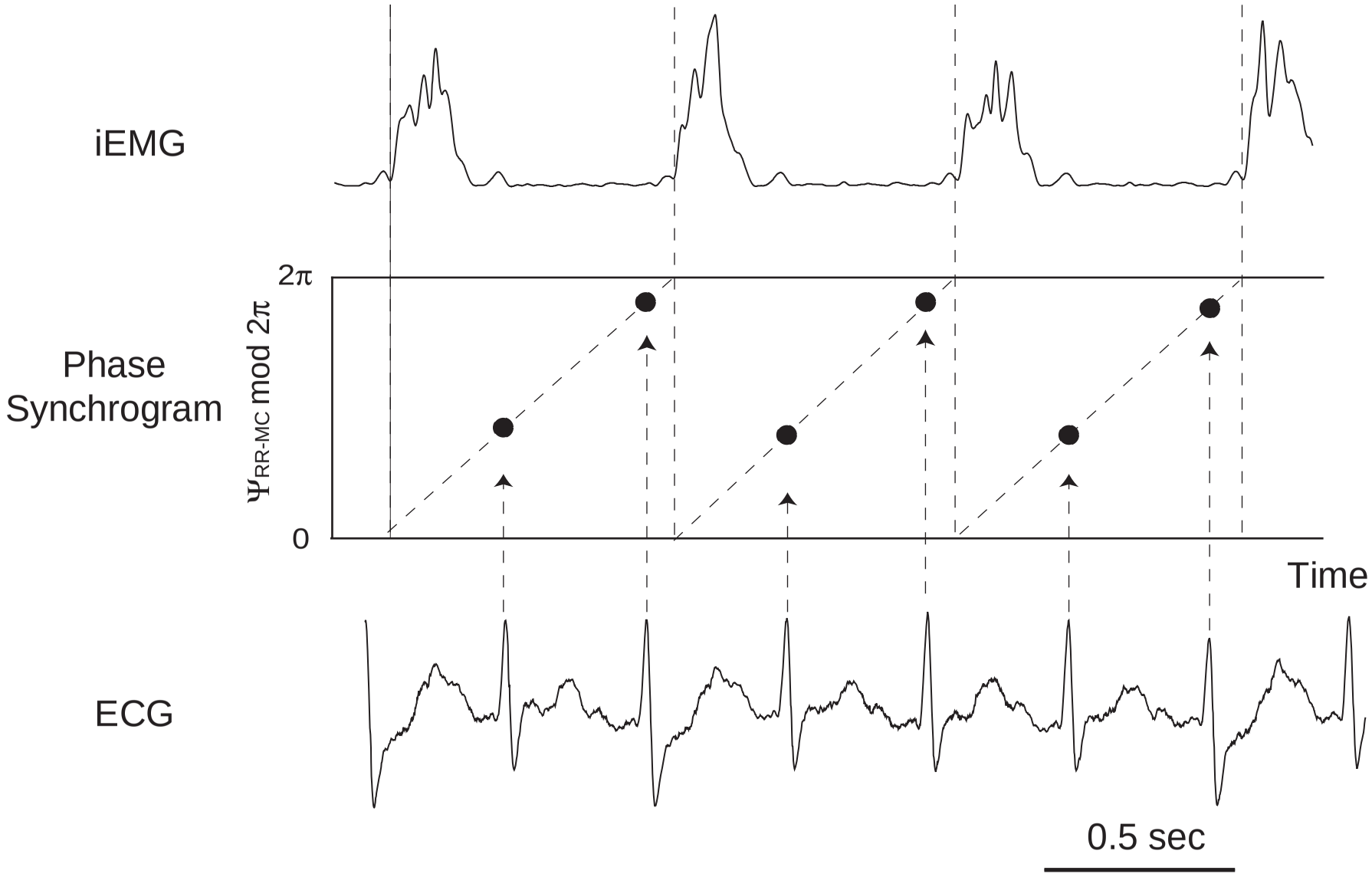
FIGURE LEGENDS

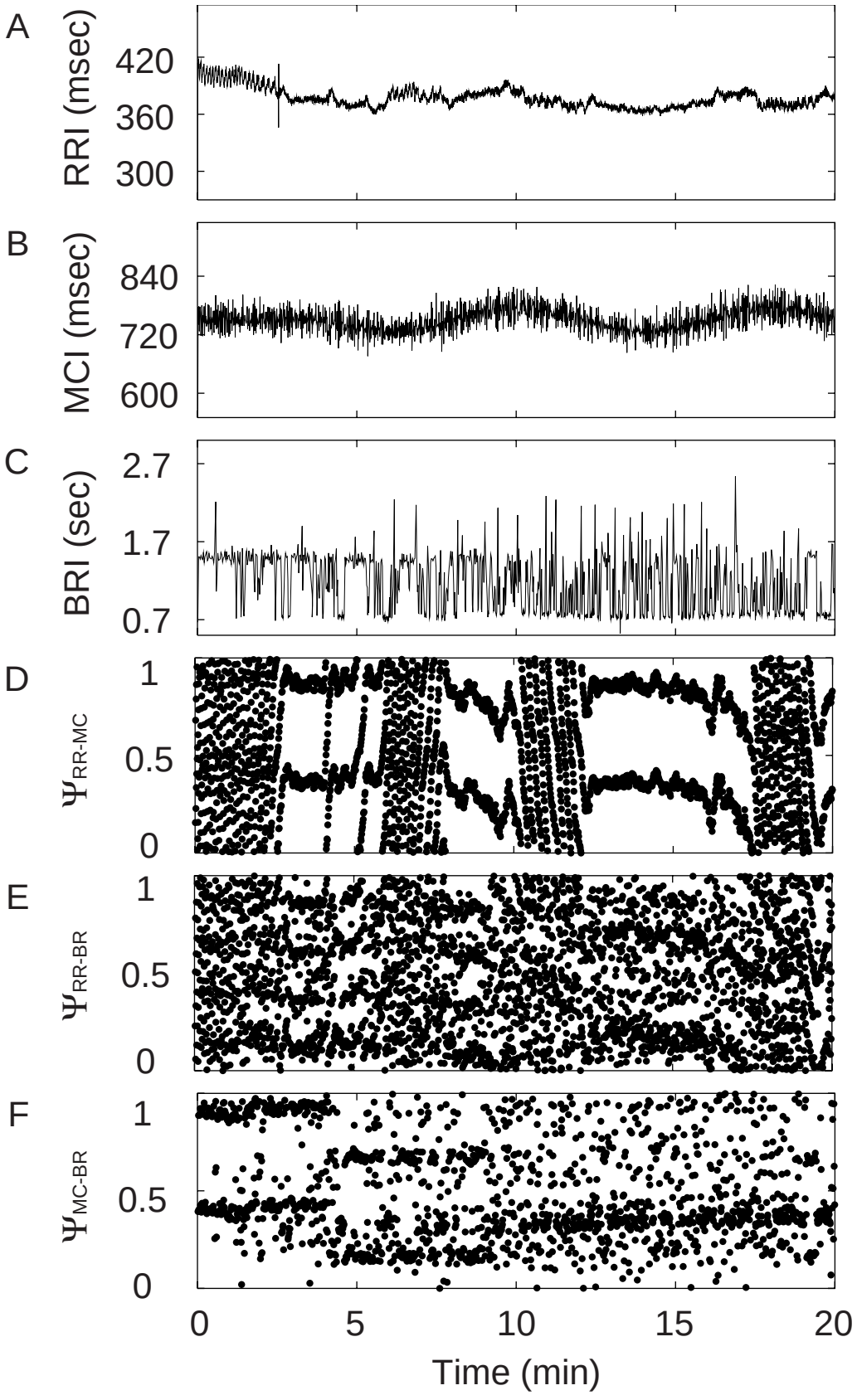
Fig. 1 The derivation of cardio-locomotor synchrogram from the generalized relative phase of locomotion (evaluated as iEMG) and the times of heart beats (R peaks in the ECG).

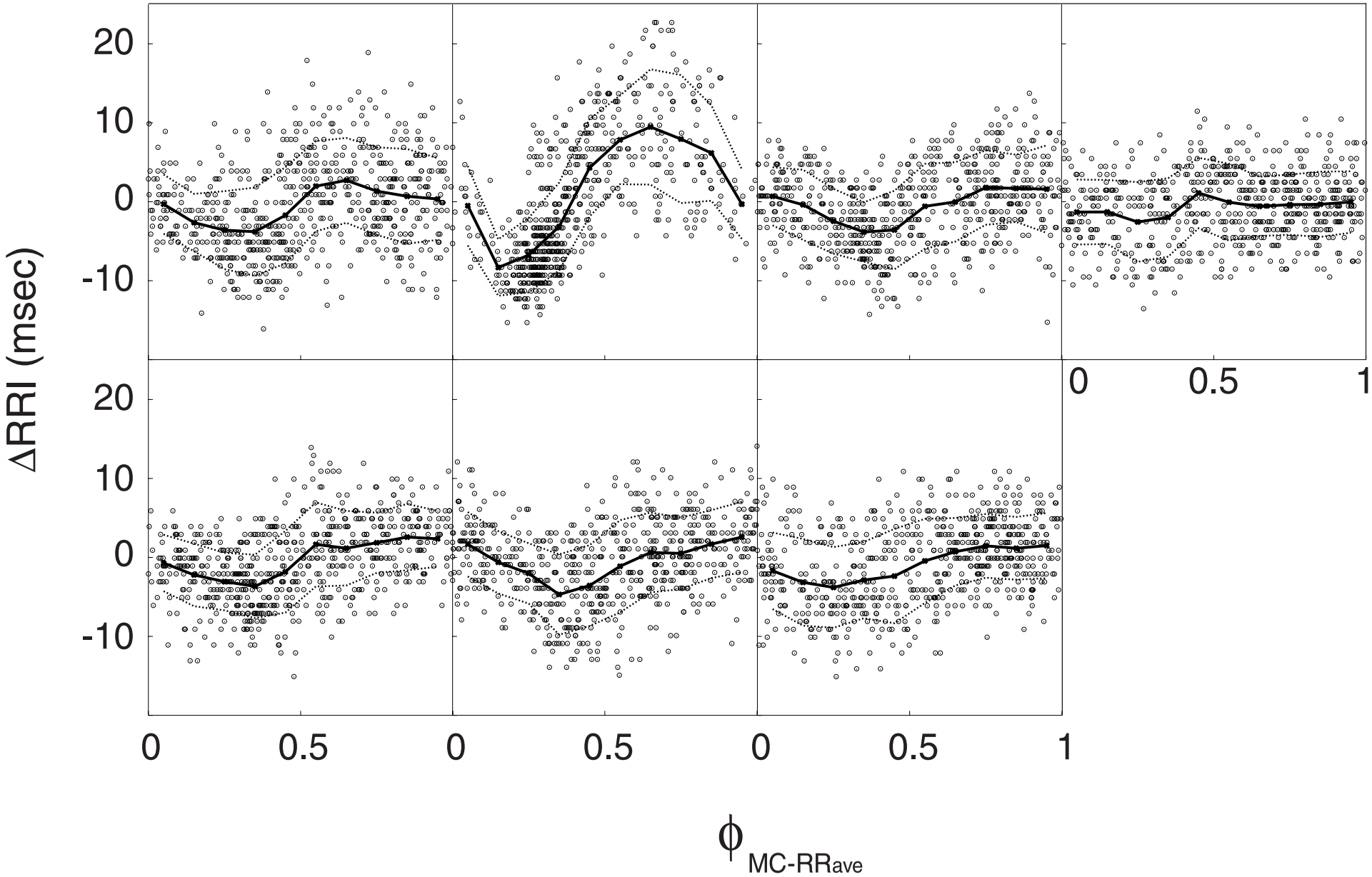
Fig. 2. Representative time series and phase synchrogram for each pair of cardiac, respiratory and locomotor rhythms. A-C: RRI, MCI, and BRI time series, respectively. Note that the MCI time series shows a sinusoidal waxing and waning after min 4 of the exercise period. D-F: Phase synchrograms between cardiac and locomotor rhythms (D), cardiac and respiratory rhythms (E), and locomotor and respiratory rhythms (F). Note that the ψ_{RR-MC} trace shows CLS even when locomotor rhythms are not constant.

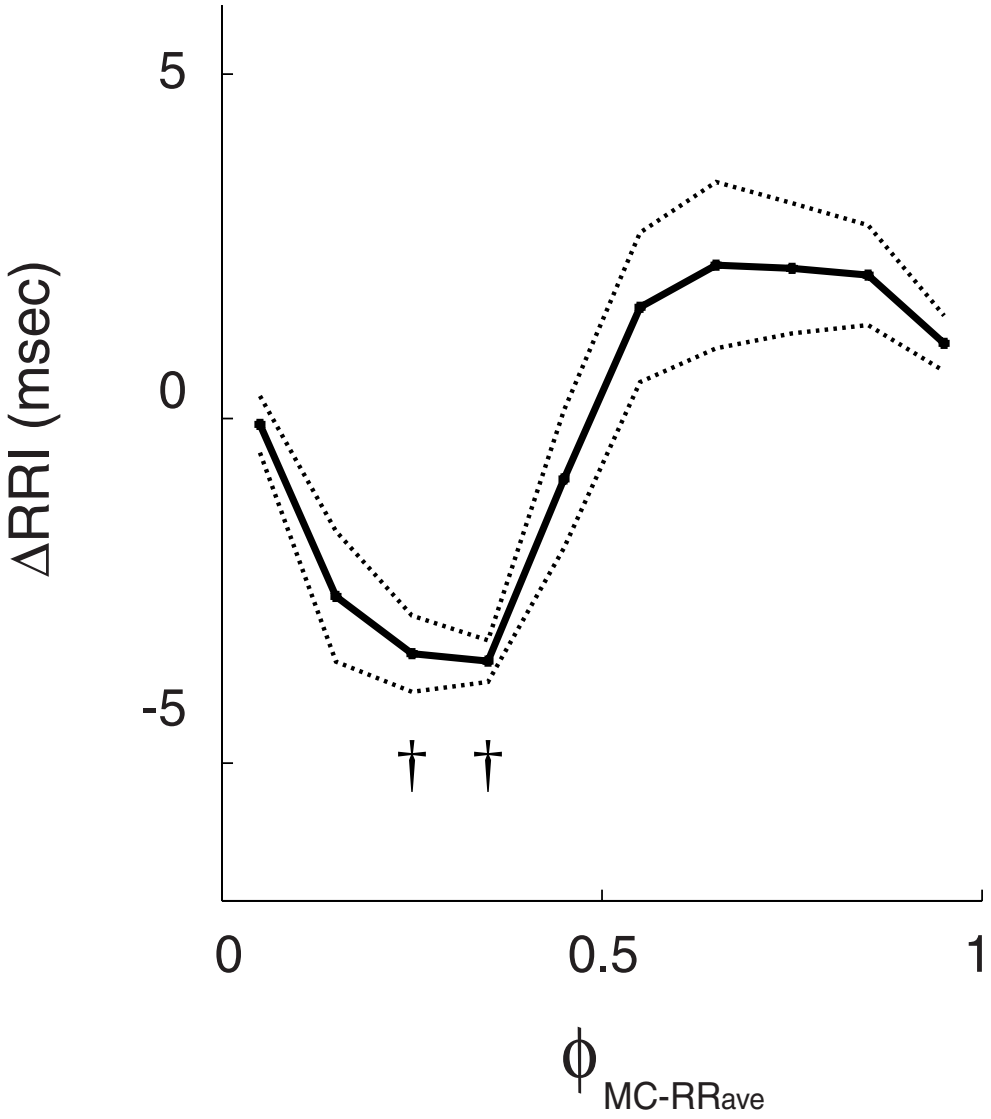
Fig. 3. Variation in RRI as a function of the phase within the cardiac cycle at which a muscle contraction occurred in all subjects. The average RRI was split into ten subintervals of equal length (classes). Solid lines indicate phase-response curve constructed by connecting mean values of the classes with line segments. The dotted line shows the standard deviation. Note that a shortening of the RRI was induced by muscle contractions that occurred within $0 < \phi_{MC-RR_{ave}} < 0.5$ and a prolongation of RRI was induced within $0.5 < \phi_{MC-RR_{ave}} < 1$.

Fig. 4. Effect of muscle contraction timing within the average RRI on the chronotropic response of the heart. The solid line shows the group mean of the phase-response curves for all subjects. The dotted line indicates the standard error. Note classes III and IV differed from class I ($P < 0.05$) This indicates a physiologicalmechanism that causes a shortening of RRI when muscle contraction occurs within the first half of the cardiac cycle.


iEMG
Phase
Synchrogram
2π
0
Ψ_{RR-MC} mod 2π
Time
ECG
0.5 sec


A
RRI (msec)
420
360
300
B
MCI (msec)
840
720
600
C
BRI (sec)
2.7
1.7
0.7
D
Ψ_{RR-MC}
1
0.5
0
E
Ψ_{RR-BR}
1
0.5
0
F
Ψ_{MC-BR}
1
0.5
0
0
5
10
15
20
Time (min)


ΔRRI (msec)
20
10
0
-10
0
0.5
1
$\phi_{MC\text{-}RRave}$


5
0
-5
ΔRRI (msec)
0
0.5
1
ϕ
MC-RRave