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Influence of time-varying mean heart rate in coronary artery disease diagnostic performance of heart rate variability indices from exercise stress testing

Raquel Bailón, PhD,[a,b,*] Pedro Serrano, MD,[c] Pablo Laguna, PhD[a,b]

[a]*Aragon Institute of Engineering Research, University of Zaragoza, Spain*
[b]*CIBER de Bioingeniería, Biomateriales y Nanomedicina (CIBER-BBN), Spain*
[c]*Department of Physiology and Pharmacology, University of Zaragoza, Spain*

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Abstract In this study, the influence of the time-varying mean heart rate (HR) and respiratory frequency in the ability of HR variability (HRV) indices to diagnose coronary artery disease has been studied. The autonomic nervous system activity has been assessed using a methodology that comprises correction of the HRV signal by the time-varying mean HR and redefinition of the classical high-frequency band to include respiratory frequencies above 0.4 Hz. The obtained clinical indices discriminate patients with coronary artery disease from patients with Framingham risk index lower than 5% with a moderate accuracy of 76%, which is lower than the reported in literature for HRV indices. We claim that time-varying mean HR and respiratory frequency, if not taken into account, introduce apparent improvement of diagnostic performance of HRV indices, adding information nonrelated to the autonomic nervous system activity, which is not what HRV is supposed to measure.

Introduction

The activity of the autonomic nervous system (ANS) on the heart is usually assessed noninvasively by means of spectral analysis of heart rate (HR) variability (HRV) that, in resting conditions, reveals the existence of at least 2 components: a low frequency (LF) component between 0.04 and 0.15 Hz and a high frequency (HF) component between 0.15 and 0.4 Hz.[1] The power in the HF band is considered to be a measure of parasympathetic activity, mainly because of respiratory sinus arrhythmia (RSA). The power in the LF band is considered to be a measure of sympathetic and parasympathetic activity together with other regulatory mechanisms such as the rennin-angiotensin system and baroreflex,[2] although its interpretation is still controversial.

Impairment of autonomic cardiovascular regulation, which causes changes in HRV, has been observed in ischemic coronary artery disease (CAD).[3,4] It has been reported that HRV is altered among patients with stable and uncomplicated CAD[3,5] and that alterations in HRV correlate with the development and severity of CAD.[3,6] Exercise stress testing (EST) is routinely used for the diagnosis of CAD. HR profile during exercise has been associated with the severity of CAD.[7] However, the value of HRV during EST in the diagnosis of CAD is controversial and has not been elucidated yet. Some studies have reported indices obtained from stress testing HRV having added value in the diagnosis of CAD.[8] On the contrary, HRV during exercise did not improve the diagnostic accuracy (AC) of traditional ST-segment analysis in the Finnish Cardiovascular study.[9]

There are 3 main concerns in the analysis of HRV during EST that, if not taken into account, may lead to erroneous interpretation of the evolution of the ANS activity: (1) local mean HR is time varying, (2) HRV during EST is highly nonstationary, and (3) respiratory frequency increases with workload and may exceed the upper limit of the classical HF band, 0.4 Hz.

The fact that local mean HR is time varying during EST makes it necessary to correct HRV before the evolution of the ANS activity is assessed from it. The reason is that mean HR influences HRV measurements as markers of autonomic function.[10] Recently, an approach for HRV analysis during EST has been proposed based on the integral pulse frequency modulation (IPFM) model, where a time-varying threshold is included to account for the time-varying mean HR.[11] The proposed approach estimates the ANS modulating signal, dividing the HRV signal by the time-varying mean HR.

* Corresponding author. Departamento de Ingeniería Electrónica y Comunicaciones, Centro Politécnico Superior, Universidad de Zaragoza, María de Luna, 1. 50018 Zaragoza, Aragón, Spain.
E-mail address: rbailon@unizar.es

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The fact that respiratory frequency is not restricted to the classical HF band during EST makes it necessary to redefine the HF band.[12] The HF band can be extended to include the whole range of possible respiratory frequencies, or it can be centered on the respiratory frequency if an estimate of it is available.

The purpose of this study is to assess whether ANS activity measured from HRV analysis during EST is capable of discriminating between patients with CAD and low CAD–risk subjects referred for EST when information on time-varying mean HR and respiratory frequency are taken into account. Our hypothesis is that the apparently promising diagnostic performance of HRV indices reported in literature is because of the inclusion of mean HR and respiratory frequency information nondirectly related to the ANS activity. The methodological approach used to assess ANS activity comprises (1) estimation of ANS modulating signal dividing the HRV signal by the time-varying mean HR, (2) nonstationary analysis of the estimated ANS modulating signal by means of the smoothed pseudo Wigner-Ville distribution (SPWVD), and (3) redefinition of the HF band to include respiratory frequencies above 0.4 Hz.

Methods and materials

Study population and acquisition

A database of exercise electrocardiograms (ECGs) of patients referred for stress testing recorded in the University Hospital Lozano Blesa of Zaragoza, Spain, was analyzed. The exercise test was performed on treadmill (Cardiovit CS-100, Schiller, Baar, Switzerland) following Bruce protocol.[13] The lead system used was the Mason-Likar modification of standard 12-lead ECG system[14] with V_2 substituted by RV_4. The usual pharmacologic treatment was not varied before any exercise test. Antianginal medications were not taken before the exercise test. The ECG leads were recorded at 1 kHz sampling rate. The investigation conformed to the principles outlined in the Declaration of Helsinki. The procedures and protocols used in this study were approved by the ethics committee of the hospital. Informed consent was obtained from all subjects before data collection.

Patients were classified into 2 different groups:

1. *Ischemic*: patients with significant stenoses in at least 1 major coronary artery, as revealed by coronary angiography (>70% narrowing of the luminal diameter).
2. *Low risk*: patients with Framingham risk index lower that 5%. Framingham risk algorithm computes the 10-year predicted risk of developing manifest CAD using data relating to several risk factors (age, total and high-density lipoprotein cholesterol, blood pressure, diabetes, and smoking).[15] Only patients with information of at least 4 risk factors were considered. Three of the patients classified as low risk by Framingham index were indeed ischemic, as revealed by coronary angiography, and were then excluded from this group. All patients in this group presented a negative clinical exercise test.

Because the purpose of the study was to analyze HRV evolution during exercise and recovery, only patients presenting no arrhythmias nor excessive number of ectopic beats, with at least 3 minutes of exercise and 3 minutes of recovery, were considered. Moreover, because one of the redefinitions of the HF band is centered on the respiratory frequency, only patients for which respiratory frequency could be estimated more than 50% of the total duration of the recording and with successive respiratory frequency estimates separated no more than 3 minutes were considered.

Some study population characteristics are shown in Table 1. More information on the database can be found in the study by Bailón et al.[8]

The IPFM model with time-varying threshold

To estimate the dynamics of the ANS activity from the available information, which are the QRS detection marks obtained in this study by the so called ARISTOTLE[16] (using leads RV_4, V_4, and V_5), the IPFM model with time-varying threshold (TVIPFM) model proposed in the study of Bailón et al[11] was used. First, an instantaneous HR signal $d_{HR}(n)$ is estimated from the QRS detection marks following a method based on the IPFM model that accounts for the presence of ectopic beats,[17] and it is resampled at a sampling rate of $F_s = 4$ Hz, then a time-varying mean HR signal $d_{HRM}(n)$ is obtained by low-pass filtering $d_{HR}(n)$ with a cutoff frequency of 0.03 Hz. The HRV signal is computed as $d_{HRV}(n) = d_{HR}(n) - d_{HRM}(n)$. Finally, the ANS modulating signal $m(n)$ is obtained, correcting the HRV signal by the time-varying mean HR, as proven in the study of Bailón et al[11] to be necessary, $m(n) = d_{HRV}(n)/d_{HR}(n)$.

Signals $d_{HR}(n)$, $d_{HRM}(n)$, and $m(n)$ are displayed in Fig. 1 for an ischemic and in Fig. 2 for a low-risk patient.

The smoothed pseudo Wigner-Ville distribution

The nonstationary analysis of the ANS modulating signal $m(n)$ was performed by means of the SPWVD[18]

$$P_m(n,\nu) = 2\sum_{k=-K+1}^{K-1} \times h(k)\left[\sum_{n'=-N+1}^{N-1} g(n')a_m(n+n'+k)a_m^*(n+n'-k)\right]e^{-j2\pi\frac{\nu}{M}k}$$

where n represents time index, $\nu = -M+1,\ldots, M$ frequency index, $a_m(n)$ is the analytic signal of $m(n)$, and $h(k)$ and $g(n)$

Table 1
Study population characteristics

Characteristic	Ischemic	Low risk
No.	52	29
Age (y)[a]	59 ± 10	40 ± 12
Sex (male/female)	50/2	20/9
MaxHR (beats/min)[a]	133 ± 19	174 ± 12
PerMaxHR (%)[a]	79 ± 18	95 ± 3
Exercise duration (min)[a]	6.2 ± 2.7	12.1 ± 13.6

MaxHR indicates maximum HR achieved; PerMaxHR, percentage of the age-predicted maximum HR achieved.

[a] Mean ± SD.

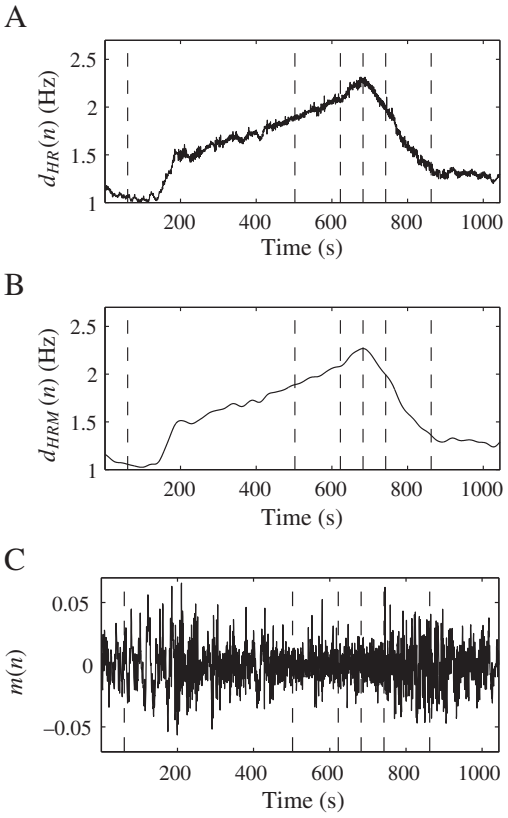

Fig. 1. Signals $d_{HR}(n)$ (A), $d_{HRM}(n)$ (B), and $m(n)$ (C) for an ischemic patient. Time instants n_1, n_{3p}, n_{1p}, n_p, n_{p1}, and n_{p3} are marked.

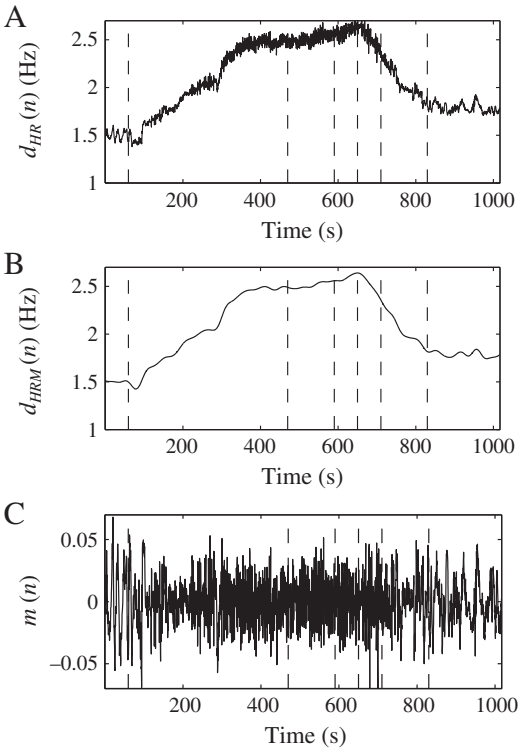

Fig. 2. Signals $d_{HR}(n)$ (A), $d_{HRM}(n)$ (B), and $m(n)$ (C) for a low-risk patient. Time instants n_1, n_{3p}, n_{1p}, n_p, n_{p1}, and n_{p3} are marked.

represent the time and frequency smoothing windows, respectively. In this study, $g(n)$ was chosen as a rectangular window of length $2N-1$ and $h(k)$ as an exponential window with damping factor γ. Used parameters values[11] are reported in Table 2.

Finally, the power of the LF and HF components were computed at each time instant n integrating $P_m(n,\nu)$ over the LF and HF bands, respectively.

Frequency bands definition

In this study, the LF band is defined in its standard way, that is, from 0.04 to 0.15 Hz, whereas 2 approaches are considered in the definition of a time-varying HF band[12]:

1. HF1: the HF band is extended to include the whole range of potential respiratory frequencies, being the upper limit given, at each time instant by half the time-varying mean HR, which is the maximum frequency with physiologic meaning.
2. HF2: the HF band is centered on the respiratory frequency, derived from the ECG using the method in the study of Bailón et al[19] and resampled at 4 Hz and has a bandwidth of 0.14 Hz.[11] The lower limit of the band is not allowed to be lower than 0.15 Hz, neither is the upper limit to be higher than half the time-varying mean HR.

Fig. 3 displays the SPWVD of the ANS modulating signal $m(n)$ for an ischemic patient, where the limits of the LF and HF (HF1 and HF2) bands are shown.

Clinical indices

To compare the dynamics of the ANS activity during EST in the ischemic and low-risk patients, the instantaneous power in the LF band, $P_{LF}(n)$, and in the HF band, considering the definitions HF1 and HF2, $P_{HF1}(n)$ and $P_{HF2}(n)$, respectively, were evaluated at different time instants: the end of the first minute of the recording (n_1), the beginning of the third minute before stress peak (n_{3p}), the beginning of the last minute before stress peak (n_{1p}), stress peak (n_p), the end of the first minute after stress peak (n_{p1}), and the end of the third minute after stress peak (n_{p3}). *Stress peak* was defined as the instant of maximum time-varying mean HR. Time-varying mean HR, $d_{HRM}(n)$, as well as the ECG-derived respiratory frequency $F_R(n)$ were also evaluated at n_1, n_{3p}, n_{1p}, n_p, n_{p1}, and n_{p3}.

Fig. 4 displays $P_{LF}(n)$, $P_{HF1}(n)$, and $P_{HF2}(n)$ for an ischemic and a low-risk patient; time instants n_1, n_{3p}, n_{1p}, n_p, n_{p1}, and n_{p3} have been marked.

Table 2
Parameter values used in the SPWVD

Parameter (U)	2M (samples)	2K−1 (samples)	2N−1 (samples)	γ (samples^{-1})
Value	1024	1023	101	1/128

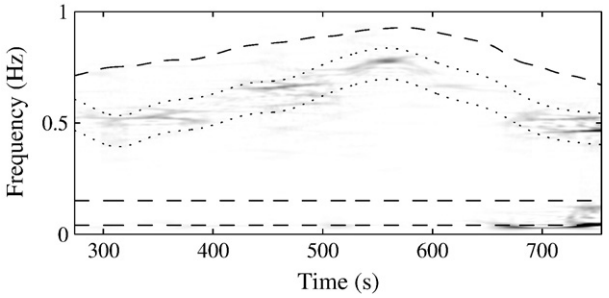

Fig. 3. Smoothed pseudo Wigner-Ville distribution of *m(n)* corresponding to an ischemic patient. Limits of the LF and HF1 bands are shown in dashed lines, and of HF2, in dotted lines.

Statistical analysis and classification

First, the nonparametric Kruskal-Wallis approach was applied to the clinical indices to compare their medians between the ischemic and low-risk patients.

Then, linear multivariate discriminant analysis was used to identify the clinical indices that best classify patients into the ischemic and low-risk groups. Feature selection was performed using the stepwise approach based on Wilk's λ minimization criterion. Classification results (sensitivity [SE], specificity [SP], positive predictive value, negative predictive value, and AC) were obtained by leave-one-out cross-validation.

Discriminant analysis assumes that variables are normally distributed. However, clinical indices based on the power of the LF and HF bands did not satisfy the normality assumption, as revealed by the Kolmogorov-Smirnov test ($P < .05$). For that reason, indices $P_{\mathrm{LF}}(n_i)$, $P_{\mathrm{HF1}}(n_i)$, and $P_{\mathrm{HF2}}(n_i)$, with $n_i \in \{n_1, n_{3p}, n_{1p}, n_p, n_{p1}, n_{p3}\}$, were logarithmically transformed before the discriminant analysis.

Results

Table 3 displays mean, SD, and median of the clinical indices in the ischemic and low-risk groups as well as those indices that were significantly different in both groups ($P < .05, .01$), as revealed by the Kruskal-Wallis test.

The power of the LF band evaluated at the first minute was significantly lower in the ischemic group with respect to the low-risk group, whereas it was significantly higher when evaluated at stress peak. The power of the HF band centered on the respiratory frequency (HF2) evaluated 3 minutes before stress peak and 1 and 3 minutes after stress peak was significantly higher in the ischemic group with respect to the low-risk group. Note that the power of the HF band extended from 0.15 Hz to half the time-varying mean HR (HF1) is higher than the power of HF2 because it may include, besides RSA, other components,[12] as depicted in Fig. 5, which displays the SPWVD of an excerpt of the ANS modulating signal corresponding to a low-risk patient. The power of HF1 evaluated at the first minute and at stress peak was significantly higher in the low-risk group with respect to the ischemic group. Respiratory frequency evaluated at 1 minute before and at stress peak was significantly lower in the ischemic group with respect to the low-risk group, as it

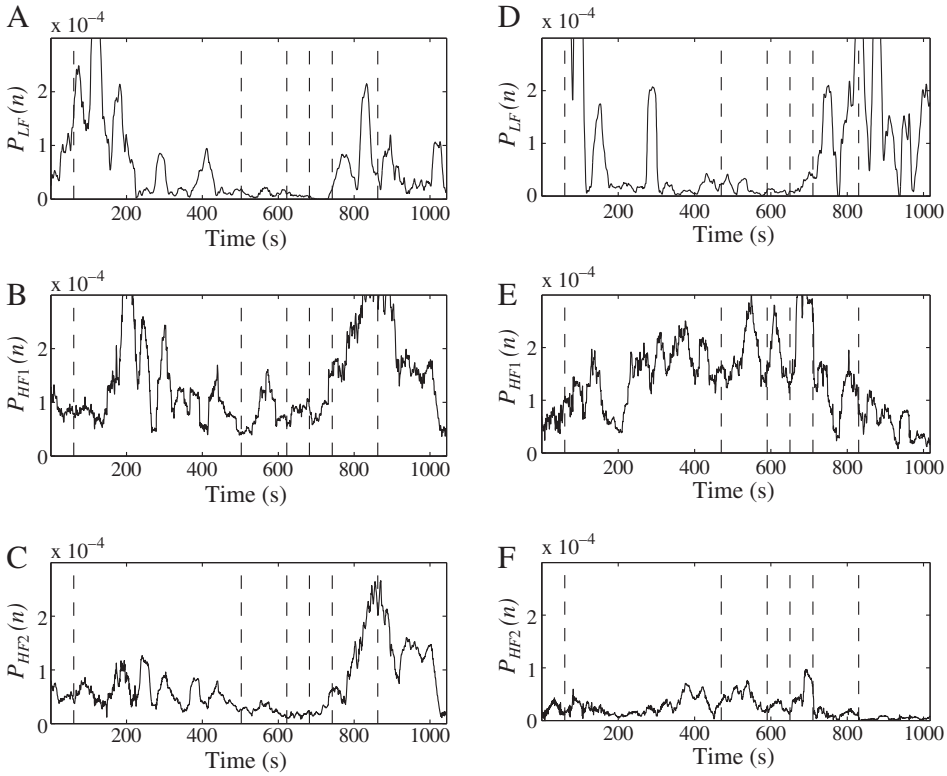

Fig. 4. $P_{\mathrm{LF}}(n)$ (A), $P_{\mathrm{HF1}}(n)$ (B), and $P_{\mathrm{HF2}}(n)$ (C) for an ischemic patient; $P_{\mathrm{LF}}(n)$ (D), $P_{\mathrm{HF1}}(n)$ (E), and $P_{\mathrm{HF2}}(n)$ (F) for a low-risk patient. Time instants n_1, n_{3p}, n_{1p}, n_p, n_{p1}, and n_{p3} are marked.

Table 3
Mean, SD, and median of clinical indices in the ischemic and low-risk groups

Clinical indices mean (SD) median	Ischemic						Low risk					
	n_1	n_{3p}	n_{1p}	n_p	n_{p1}	n_{p3}	n_1	n_{3p}	n_{1p}	n_p	n_{p1}	n_{p3}
$P_{LF}(n_i)$ ($\times 10^6$)	609.8	61.2	38.2	179.4	251.6	657.2	1145.3	19.7	9.1	4.8	68.0	784.9
	(1161.0)	(120.1)	(89.7)	(927.8)	(415.3)	(1077.2)	(1332.7)	(21.3)	(9.6)	(4.0)	(71.6)	(1157.5)
	160.4*	18.2	8.9	9.4*	68.9	231.3	646.9*	9.9	5.5	4.8*	36.6	400.5
$P_{HF1}(n_i)$ ($\times 10^6$)	242.2	103.7	79.5	85.5	242.0	374.0	635.5	65.4	102.8	146.2	97.9	173.1
	(286.1)	(121.2)	(62.4)	(80.5)	(287.5)	(568.6)	(908.8)	(45.5)	(67.0)	(155.8)	(64.7)	(223.8)
	130.9*	61.9	61.7	55.5*	116.6	208.0†	270.1*	52.6	85.4	115.8*	80.1	87.6†
$P_{HF2}(n_i)$ ($\times 10^6$)	109.0	33.6	23.1	23.4	77.3	151.5	371.8	11.1	13.2	12.5	24.1	45.6
	(140.5)	(38.1)	(31.6)	(29.6)	(113.8)	(240.4)	(714.6)	(10.4)	(10.8)	(8.9)	(25.8)	(70.5)
	44.1†	22.8*	13.2	14.4	33.8*	75.9*	114.0†	7.5*	10.1	10.5	11.9*	27.0*
$F_R(n)$ (Hz)	0.30	0.39	0.45	0.48	0.46	0.40	0.29	0.45	0.55	0.57	0.52	0.43
	(0.09)	(0.10)	(0.12)	(0.12)	(0.10)	(0.08)	(0.08)	(0.13)	(0.12)	(0.12)	(0.11)	(0.13)
	0.29	0.39†	0.44*	0.47*	0.45†	0.39	0.29	0.43†	0.55*	0.59*	0.53†	0.44
d_{HRM} (n_i) (Hz)	1.4	1.9	2.1	2.2	1.9	1.5	1.5	2.5	2.7	2.9	2.5	1.8
	(0.3)	(0.3)	(0.3)	(0.3)	(0.3)	(0.3)	(0.3)	(0.3)	(0.3)	(0.2)	(0.3)	(0.2)
	1.4	1.9*	2.1*	2.2*	1.9*	1.5*	1.4	2.5*	2.7*	2.9*	2.5*	1.8*

† $P < .05$.
* $P < .01$.

happened with the time-varying mean HR evaluated at every time instant except for the first minute.

Linear multivariate discriminant analysis was applied independently to different subsets of features: (1) ANS-related indices, that is, the power in the LF and HF band (HF1 and HF2 were studied separately); (2) respiratory frequency indices; (3) time-varying mean HR indices; and (4) all clinical indices. Classification results are displayed in Table 4. Classification features used in each case are listed in the same order as selected by the stepwise method together with the standardized canonical discriminant function coefficients.

Discussion

Methodological aspects

The shortcomings of extending interpretation of classical HRV analysis at rest to the setting of dynamic exercise have been already pointed out[2] and may explain the differences found in different studies.[20]

In this manuscript, the dynamics of the ANS activity during EST have been assessed using a method that comprises (1) estimation of ANS modulating signal based on the TVIPFM model, which takes into account the time-varying mean HR; (2) nonstationary analysis based on the SPWVD; and (3) redefinition of the HF band to include respiratory frequencies above 0.4 Hz.

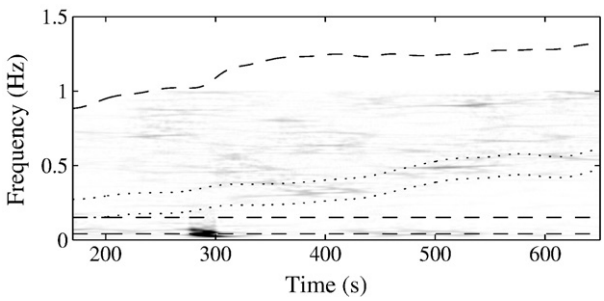

Fig. 5. Smoothed pseudo Wigner-Ville distribution of $m(n)$ during the exercise phase corresponding to a low-risk patient. Limits of the LF and HF1 bands are shown in dashed lines, and of HF2, in dotted lines.

It has been shown that mean HR influences HRV parameters as markers of autonomic activity.[9-11,21] For example, if the power of the LF and HF components are obtained from the HR signal (heart period signal), an increase in mean HR leads to an overestimated (underestimated) ANS modulating signal.[11] This justifies the necessity of correcting the HRV signal during EST with the time-varying mean HR before the evolution of the ANS activity is assessed from it. This is extremely important in our study not only because the mean HR is time-varying during EST but also because mean HR during exercise and recovery is significantly higher in the low-risk group with respect to the ischemic group (Table 3).

To overcome the limitation that respiratory frequency during EST may exceed the upper limit of the classical HF band, 0.4 Hz, 2 approaches have been considered in this study. Approach HF2 requires the knowledge of respiratory frequency, which can be obtained either from a simultaneously recorded respiratory signal or from an ECG-derived respiratory signal using, for example, as in this study, the method described in the study of Bailón et al.[19] This method is not applicable in some situations, for example, when respiratory signal does not exhibit a unimodal pattern (then it does not make sense to speak about respiratory frequency) or when HR is lower than twice the respiratory frequency (then aliased components appear). On the other hand, approach HF1 does not rely on any prior information but may include not only the RSA but also other HF components. In our study population, power in HF1 was significantly higher ($P < .001$) than in HF2 for all time instants considered, which suggests the presence in HF1 of other components besides RSA (Fig. 5).

CAD diagnostic performance of the clinical indices was evaluated using linear multivariate discriminant analysis. However, other classification methods, such as support

Table 4
Classification results: ischemic vs low-risk groups

Feature subset Classification variables[a] (Standardized coefficients)[b]	SE (%)	SP (%)	$^{+}$P (%)	$^{-}$P (%)	AC (%)
ANS-related indices (HF1) $P_{LF}(n_p)$, $P_{HF1}(n_p)$, $P_{LF}(n_1)$, $P_{HF1}(n_{p3})$, $P_{HF1}(n_{3p})$ (0.623, −1.015, −0.618, 0.518, 0.539)	85	90	94	76	86
ANS-related indices (HF2) $P_{HF2}(n_{3p})$, $P_{LF}(n_1)$, $P_{LF}(n_p)$, $P_{HF2}(n_{p3})$ (0.605, −0.606, 0.431, 0.433)	75	79	87	64	76
Respiratory frequency indices $F_R(n_{1p})$ (1)	75	72	83	62	74
Time-varying mean HR indices $d_{HRM}(n_p)$, $d_{HRM}(n_1)$ (1.135, −0.454)	88	93	96	82	90
All clinical indices (HF1) $d_{HRM}(n_p)$, $d_{HRM}(n_1)$, $P_{HF1}(n_p)$, $F_R(n_{p1})$ (1.084, −0.397, 0.338, 0.334)	96	93	96	93	95
All clinical indices (HF2) $d_{HRM}(n_p)$, $d_{HRM}(n_1)$, $F_R(n_{p3})$ (1.153, −0.497, 0.330)	88	90	94	81	89

$^{+}$P indicates positive predictive value; $^{-}$P, negative predictive value.
[a] Listed in the same order as selected by the stepwise method.
[b] Standardized canonical discriminant function coefficients.

vector machine, may obtain better classification results than discriminant analysis.

Physiologic aspects

In this study, the dynamics of the ANS activity during EST have been compared in 2 groups of patients referred for treadmill EST: ischemic and low-risk.

The power in the LF band evaluated at the first minute of the recording was significantly lower in the ischemic group with respect to the low-risk group, whereas it was significantly higher when evaluated at stress peak. Regarding HF band, power in HF2 evaluated at the first minute of exercise was significantly lower in the ischemic group, but significantly higher power was observed in the ischemic group with respect to the low-risk group when evaluated 3 minutes before stress peak and 1 and 3 minutes after stress peak. In the study by Virtanen et al,[9] the SD of R-R intervals standard deviation of the normal to normal interval (SDNN), which has been shown to correlate with total power, as well as the square root of the mean-squared differences of successive R-R intervals squared root of the mean squared differences of successive normal to normal intervals (RMSSD), previously shown to correlate with the HF power, were significantly lower in patients with CAD during the first minute of exercise, which is in agreement with our results. However, SDNN and RMSSD during the last minute of exercise were significantly lower in patients with CAD, which is in contrast with our results. During recovery, the only significant difference between patients with CAD and patients who are non-CAD was RMSSD during the second minute. The disagreement with our results may be due to the comparison between time and frequency domain indices, to the different correction applied either to R-R intervals or HRV signal before computing HRV indices, or to the redefinition of HF2. In fact, if the power in HF1 is considered, it turns out that, at stress peak, it was significantly higher in the low-risk group. In the study by Dilaveris et al,[22] a significant decrease in parasympathetic activity during exercise and an increase during the recovery period were reported both in patients with CAD and in healthy controls as well as a predominance of sympathetic tone during the recovery, which is in concordance with our results. However, the sympathetic predominance during recovery was more pronounced in patients with CAD than in healthy controls, which is not observed in our study. The fact that in the study by Dilaveris et al[22] the standard HF band (0.15-0.4 Hz) was used may explain the apparent differences.

There are other studies that have reported reduced LF and HF power in patients with CAD with respect to patients who are non-CAD and healthy volunteers,[5,6] but they analyzed HRV either during rest or 24-hour recordings, not during exercise.

Indices related to the ANS activity (HF2) obtained a discrimination AC of 76%, which is similar to the one reported in the study by Virtanen et al,[9] although different HRV indices were used. When classification performance of ANS-related indices was assessed, using the power in HF1 instead of in HF2 increased both SE and SP (from 75% to 85% and from 79% to 90%, respectively). In the study by Bailón et al,[8] classical HRV frequency indices obtained an AC over a database that comprises our study population of 89% and up to 93% when HRV time and frequency indices were combined. However, there is a limitation in the interpretation of those indices as ANS activity markers. First, because the influence of the mean HR was not corrected, those indices may include information on the mean HR changes during EST, which, as displayed in Table 4, achieves a classification AC of 90%, then classical HF band (0.15-0.4 Hz) was used in conjunction with a new band, the very HF band, that extended from 0.4 to 1 Hz. In Table 3, it can be observed that respiratory frequency exceeds the upper limit of the classical HF band during exercise and recovery in both groups, making the interpretation of the classical HF and very HF bands power difficult. Finally, one of the classification variables that was included and selected in the analysis was the slope of the linear detrending of the HRV signal in each analyzed period, which, in fact, is accounting for the changes in the mean HR that, as previously stated, showed high-classification AC.

The classification performance of the respiratory frequency and the time-varying mean HR was also evaluated. Respiratory frequency 1 minute before stress peak obtained a classification AC of 74%, which may also be related to the different workloads supported by the ischemic and low-risk groups. A combination of the mean HR at the first minute and at stress peak obtained a SE of 88% and an SP of 93%, and when information of the power in HF1 and respiratory frequency is included, SE increased to 93%. However, the power in HF2 did not add any classification improvement to the time-varying mean HR indices. The high-classification AC achieved by the time-varying mean HR indices is in agreement with Serkan et al,[7] where it was shown that

decreased HR increment and decreased HR decrement during exercise and recovery is strongly associated with the severity of CAD. However, in our study population, the classification AC of time-varying mean HR indices may be justified by the difference in mean ages between the ischemic and low-risk groups as well as the negative chronotropic drugs taken by some patients.

The origin of the HF components besides the RSA observed in HF1 is uncertain. They may be related to vagal reaction or fragmented respiration or they may be unrelated to the parasympathetic system activity. Our hypothesis is that those components may have a similar origin than the pedaling component, sometimes present in EST performed on bicycle ergometer,[23,24] that originates from a dynamic modulation of venous return by rhythmical limb muscles contractions and whose power increases with workload.[23] The venous return modulation is likely to induce a rhythmical stretching of the sinus node that modulates HR. There are some works in running subjects reporting a blood pressure modulation at the stride rate,[25] which may be the origin of those HF components. If our hypothesis is true, the increase in classification AC obtained by HF1 with respect to HF2 may be justified by the higher workloads supported by the low-risk group, as revealed by the minutes of exercise and the percentage of the theoretical maximum HR achieved in the EST (Table 1).

Limitations

One limitation of this study is that there exists a bias in the study population because the traditional interpretation of EST influences the decision to perform a coronary angiography. This limitation is unavoidable in studies that use coronary angiography as a reference standard because it is unviable to perform a coronary angiography to all patients independently of the EST results. Other limitations are the unbalanced number of men and women and the relatively small number of patients in the study.

Conclusions

In this study, the effect of the time-varying mean HR and respiratory frequency in the CAD diagnostic performance of HRV indices has been studied. The ANS activity has been assessed using a methodology that comprises correction of the HRV signal by the time-varying mean HR and redefinition of the classical HF band to include respiratory frequencies above 0.4 Hz.

Indices related to the ANS activity (power in the LF band and RSA) discriminate patients with CAD from patients with Framingham risk index lower than 5% with a moderate AC of 76%, which is lower than the reported in literature for HRV indices. The diagnostic AC increased to 86% when other HF components besides the RSA were considered and reached 95% when indices related to mean HR were considered (although, a 90% was already achieved using only the mean HR at the first minute and at stress peak). The main conclusion of this work is that care should be taken in the interpretation of HRV parameters and their diagnostic performance if information on the time-varying mean HR and respiratory frequency are not taken into account. Otherwise, the diagnostic performance of HRV indices may be due to differences in the time-varying mean HR and respiratory frequency rather than to differences in the ANS activity.

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References

1. The Task Force of ESC and NASPE. Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Eur Heart J 1996;17:354.
2. Aubert A, Seps B, Beckers F. Heart rate variability in athletes. Sports Med 2003;33:889.
3. Huikuri HV, Mäkikallio TH. Heart rate variability in ischemic heart disease. Auton Neurosci 2001;90:95.
4. Sroka K, Peimann C, Seevers H. Heart rate variability in myocardial ischemia during daily life. J Electrocardiol 1997;30:45.
5. Wennerblom B, Lurje L, Tygesen H, Vahisalo R, Hjalmarson A. Patients with uncomplicated coronary artery disease have reduced heart rate variability mainly affecting vagal tone. Heart 2000;83: 290.
6. Hayano J, Sakakibara Y, Yamada A, et al. Decreased magnitude of heart rate spectral components in coronary artery disease. Its relation to angiographic severity. Circulation 1990;81:1217.
7. Serkan C, Ozturk S, Biyikoglu F, et al. Association of heart rate profile during exercise with the severity of coronary artery disease. J Cardiovasc Med 2009;10:394.
8. Bailón R, Mateo J, Olmos S, et al. Coronary artery disease diagnosis based on exercise electrocardiogram indexes from repolarisation, depolarisation and heart rate variability. Med Biol Eng Comput 2003;41:561.
9. Virtanen M, Kähönen M, Nieminen T, et al. Heart rate variability derived from exercise ECG in the diagnosis of coronary artery disease. Physiol Meas 2007;28:1189.
10. Chiu H, Wang T, Huang L, Tso H, Kao T. The influence of mean heart rate on measures of heart rate variability as markers of autonomic function: a model study. Med Eng Phys 2003;25:475.
11. Bailón R, Laouini G, Grao C, Orini M, Laguna P, Meste O. The integral pulse frequency modulation model with time-varying threshold: application to heart rate variability analysis during exercise stress testing. IEEE Trans Biomed Eng 2011, in press.
12. Bailón R, Laguna P, Mainardi L, Sörnmo L. Analysis of heart rate variability using time varying frequency bands based on respiratory frequency. In Proc 29th Int Conf IEEE Eng Med Biol Soc 2007: 6674.
13. Bruce RA, Kusumi F, Hosmer D. Maximal oxygen intake and nomographic assessment of functional aerobic impairment in cardiovascular disease. Am Heart J 1973;85:546.
14. Mason RE, Likar I. A new system of multiple-lead exercise electrocardiography. Am Heart J 1966;71:196.
15. D'Agostino R, Russell M, Muse D, Ellison C, Silbershatz H, Wilson P. Primary and subsequent coronary risk appraisal: new results from the Framingham study. Am Heart J 2000;139:272.
16. Moody GB, Mark RG. Development and evaluation of a 2-lead ECG analysis program. In Proc Comput Cardiol 1982:39.
17. Mateo J, Laguna P. Analysis of heart rate variability in the presence of ectopic beats using the heart timing signal. IEEE Trans Biomed Eng 2003;50:334.

18. Bailón R, Mainardi L, Orini M, Sörnmo L, Laguna P. Analysis of heart rate variability during exercise stress testing using respiratory information. Biomed Signal Process Control 2010;5:299.
19. Bailón R, Sörnmo L, Laguna P. A robust method for ECG based estimation of the respiratory frequency during stress testing. IEEE Trans Biomed Eng 2006;53:1273.
20. Cottin F, Papelier Y. Regulation of cardiovascular system during dynamic exercise: integrative approach. Crit Rev Physical Rehab Med 2002;14:53.
21. Sacha J, Pluta W. Different methods of heart rate variability analysis reveal different correlations of heart rate variability spectrum with average heart rate. J Electrocardiol 2005;38:47.
22. Dilaveris P, Zervopoulos G, Michaelides A, et al. Ischemia-induced reflex sympathoexcitation during the recovery period after maximal treadmill exercise testing. Clin Cardiol 1998;21:585.
23. Blain G, Meste O, Blain A, Bermon S. Time-frequency analysis of heart rate variability reveals cardiolocomotor coupling during dynamic cycling exercise in humans. Am J Physiol Heart Circ Physiol 2009; 296:1651.
24. Villa F, Castiglioni P, Merati G, Mazzoleni P, Di Rienzo M. Effects of pedalling on the high frequency components of HRV during exercise. In Proc Comput Cardiol 2008;35:37.
25. Palatini P, Mos L, Mormino P, et al. Blood pressure changes during running in humans: the “beat” phenomenon. J Appl Physiol 1989;67:52.