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Kubios HRV – Heart rate variability analysis software

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ABSTRACT

Kubios HRV is an advanced and easy to use software for heart rate variability (HRV) analysis. The software supports several input data formats for electrocardiogram (ECG) data and beat-to-beat RR interval data. It includes an adaptive QRS detection algorithm and tools for artifact correction, trend removal and analysis sample selection. The software computes all the commonly used time-domain and frequency-domain HRV parameters and several nonlinear parameters. There are several adjustable analysis settings through which the analysis methods can be optimized for different data. The ECG derived respiratory frequency is also computed, which is important for reliable interpretation of the analysis results. The analysis results can be saved as an ASCII text file (easy to import into MS Excel or SPSS), Matlab MAT-file, or as a PDF report. The software is easy to use through its compact graphical user interface. The software is available free of charge for Windows and Linux operating systems at http://kubios.uef.fi.

1. Introduction

Heart rate variability (HRV) analysis is generally used for evaluating autonomic nervous system (ANS) functioning in cardiovascular research and in different human wellbeing related applications. HRV is known to be affected, e.g. by stress, certain cardiac diseases and pathological states. HRV is a result of ANS regulation of the sinoatrial (SA) node. ANS is divided into sympathetic and parasympathetic branches and their influences on heart rate (HR) and HRV are quite well understood. Roughly speaking, sympathetic activity tends to increase HR and decrease HRV, whereas parasympathetic tends to decrease HR and increase HRV [1]. The most conspicuous periodic component of HRV is the respiratory sinus arrhythmia (RSA) which is considered to range from 0.15 to 0.4 Hz. This high frequency (HF) component is mediated almost solely by the parasympathetic nervous activity [1,2]. Another apparent component of HRV is the low frequency (LF) component ranging from 0.04 to 0.15 Hz. The LF component is generally thought of being both of sympathetic and parasympathetic origin [1], but there are studies demonstrating that the normalized value of the LF component could be used to assess sympathetic efferent activity [3,4].

One of the main clinical scenarios where HRV has been found valuable include the risk stratification of sudden

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cardiac death after acute myocardial infarction [5,6,2]. In addition, HRV is generally accepted to provide an early warning sign of diabetic neuropathy [5,6]. Besides these main clinical scenarios, HRV has been studied with relation to several cardiovascular diseases, renal failure, physical exercise, occupational and psychosocial stress, gender, age, drugs, alcohol, smoking and sleep [7,8,5,9,10,6].

The popularity of HRV analysis has lead to rise of several commercial and non-commercial software tools. Many commercial electrocardiography (ECG) and HR monitor devices include software for HRV analysis, but there are also few commercial device-independent HRV analysis software tools. In addition to commercial tools, several non-commercial HRV software tools have been developed. These include a Matlab based software package called POLYAN [11], a Matlab toolbox called ECGLab [12], another Matlab based software called KARDIA [13], the HRV toolkit available at PhysioNet [14], an R language based software package called RHRV [15], and a software tool for HRV, T-wave alternans and heart rate turbulence analysis [16].

In this paper, we introduce the Kubios HRV software (ver. 2.1) which is an easy to use HRV analysis tool including a wide variety of time-domain, frequency-domain and nonlinear analysis options. The very first version of Kubios HRV (ver. 1.1) was published in 2004 [17] and a considerable upgrade (ver. 2.0) with extra options for nonlinear analysis and improved usability was released in 2008 [18]. Kubios HRV has become a popular analysis tool for HRV which is attested by over 16 000 downloads of the latest version (ver. 2.0) within the past four years that it has been distributed. The new version (ver. 2.1) is a significant upgrade of the previous version including electrocardiogram (ECG) data support, built-in QRS detector accompanied by artifact correction tools, respiratory frequency estimation (ECG derived respiration, EDR), and several usability and functionality improvements. The software is available free of charge for Windows and Linux operating systems at http://kubios.uef.fi.

2. Computational methods

This section describes shortly the analysis parameters included in Kubios HRV. The computations as well as the notations used are mainly based on the guidelines given in [5]. Details of different parameters can be found from the references given below or from the Kubios HRV User's Guide (http://kubios.uef.fi). A summary of the analysis parameters is given in Table 1.

2.1. QRS detection algorithm

In case ECG data is available, the R-wave time instants are automatically detected by applying a built-in QRS detection algorithm. This in-house developed detection algorithm is based on the Pan–Tompkins algorithm [29]. The detector consists of a preprocessing part followed by decision rules. The preprocessing part includes bandpass filtering of the ECG (to reduce power line noise, baseline wander and other noise components), squaring of the data samples (to highlight peaks) and moving average filtering (to smooth close-by peaks). The decision rules include amplitude threshold and expected time between adjacent R-waves. Both of these rules are adjusted adaptively every time a new R-wave is detected. Before R-wave time instant extraction, the R-wave is interpolated at 2000 Hz to improve the time resolution of the detection. The upsampling can significantly improve the time resolution of R-wave detection when the sampling rate of the ECG is low.

2.2. Time-domain methods

The time-domain methods are computationally simple and they are applied directly to the series of successive RR interval values [5]. The most evident such measure is the mean value of RR intervals ($\overline{RR}$) or, correspondingly, the mean HR ($\overline{HR}$). In addition, several variables that measure the variability within the RR series exist. The standard deviation of normal-to-normal RR intervals (SDNN) reflects the overall (both short-term and long-term) variation within the RR interval series, whereas the root mean square of successive differences (RMSSD) can be used as a measure of the short-term variability. Another measure calculated from successive RR interval differences is the NN50 which is the number of successive intervals differing more than 50 ms or the corresponding relative amount pNN50.

Other time-domain parameters computed from successive 5-min segments include the standard deviation of the average RR intervals calculated over the 5-min segments (SDANN) and the mean of the 5-min standard deviations of RR intervals (SDNN index) [5]. The computation of SDANN and SDNN index require long-term measurements, basically 24-h Holter recordings.

In addition to the above statistical parameters, there are two geometric measures which are calculated from the RR interval histogram. The HRV triangular index is obtained as the integral of the histogram (i.e. total number of RR intervals) divided by the height of the histogram which depends on the selected bin width. In order to obtain comparable results, a bin width of 1/128 s is used as recommended in [5]. Another geometric measure is TINN which is the baseline width of the RR histogram evaluated through triangular interpolation [5].

2.3. Frequency-domain methods

In the frequency-domain methods, a spectrum estimate is calculated for the RR interval series. Prior to spectrum estimation, the RR interval series is converted to equidistantly sampled series by cubic spline interpolation. In the software, the spectrum is estimated with two different methods: Welch's periodogram and autoregressive (AR) modelling. In Welch's periodogram, the RR series is divided into overlapping segments, each segment is windowed to decrease the leakage effect, and the spectrum estimate is obtained by averaging the FFT spectra of these windowed segments. In the AR method, the RR series is modelled with an AR model of specific order and the spectrum estimate is obtained from the estimated model parameters. The AR spectrum can be divided into distinct spectral components by applying spectral factorization [30].

Table 1 – Summary of HRV parameters calculated by Kubios HRV software. The frequency-domain parameters (all except the EDR) are computed using two different spectrum estimation methods described in the text.

Parameter	Units	Description	References
Time-Domain			
$\overline{RR}$	[ms]	The mean of RR intervals	[5]
STD RR (SDNN)	[ms]	Standard deviation of RR intervals	–″–
$\overline{HR}$	[1/min]	The mean heart rate	–″–
STD HR	[1/min]	Standard deviation of instantaneous heart rate values	–″–
RMSSD	[ms]	Square root of the mean squared differences between successive RR intervals	–″–
NN50	[count]	Number of successive RR interval pairs that differ more than 50 ms	–″–
pNN50	[%]	NN50 divided by the total number of RR intervals	–″–
HRV triangular index	–	The integral of the RR interval histogram divided by the height of the histogram	–″–
TINN	[ms]	Baseline width of the RR interval histogram	–″–
Frequency-Domain			
VLF, LF and HF peaks	[Hz]	Peak frequencies for VLF, LF and HF bands	[5]
VLF, LF and HF powers	[ms^2]	Absolute powers of VLF, LF and HF bands	–″–
VLF, LF and HF powers	[%]	Relative powers of VLF, LF and HF bands	–″–
		VLF [%] = VLF [ms^2]/total power [ms^2] × 100 %	–″–
		LF [%] = LF [ms^2]/total power [ms^2] × 100 %	–″–
		HF [%] = HF [ms^2]/total power [ms^2] × 100 %	–″–
LF and HF powers	[n.u.]	Powers of LF and HF bands in normalized units	–″–
		LF [n . u .] = LF [ms^2]/(total power [ms^2] − VLF [ms^2])	–″–
		HF [n . u .] = HF [ms^2]/(total power [ms^2] − VLF [ms^2])	–″–
LF/HF	–	Ratio between LF and HF band powers	–″–
Total power	[ms^2]	Total spectral power	–″–
EDR	[Hz]	ECG derived respiratory frequency	[19]
Nonlinear			
SD1, SD2	[ms]	Standard deviations of the Poincaré plot	[20]
ApEn	–	Approximate entropy	[21,22]
SampEn	–	Sample entropy	[21]
D_2	–	Correlation dimension	[23,24]
α_1, α_2	–	Short-term and long-term fluctuations of detrended fluctuation analysis (DFA)	[25,26]
Lmean	[beats]	Mean line length of diagonal lines in recurrence plot (RP)	[27,28]
Lmax	[beats]	Maximum line length of diagonal lines in RP	–″–
REC	[%]	Recurrence rate (percentage of recurrence points in RP)	–″–
DET	[%]	Determinism (percentage of recurrence points which form diagonal lines in RP)	–″–
ShanEn	–	Shannon entropy of diagonal line lengths' probability distribution	–″–

The spectrum estimates are then divided into very low frequency (VLF), low frequency (LF), and high frequency (HF) bands. The generally used limits for these bands in case of short-term HRV recordings in normal human subjects are 0–0.04 Hz (VLF), 0.04–0.15 Hz (LF) and 0.15–0.4 (HF). HRV measures extracted from these frequency bands include peak frequencies (i.e. the frequency values corresponding to maximum power within VLF, LF, and HF bands), absolute and relative powers (for VLF, LF, and HF), normalized powers (for LF and HF), LF/HF power ratio, and the total spectral power. The band powers are computed by simply integrating the spectrum estimates over the frequency band limits, and total power by integrating over the whole spectrum. In the case of AR spectrum, when the spectral factorization is used, the power of a specific frequency band is obtained by summing the powers of components within the band. The computation of the relative and normalized powers are given in Table 1.

Because the RSA (i.e. the HF) component of HRV is centred around the respiratory frequency, the respiration should always be considered in HRV analysis. This is because the respiratory frequency varies between subjects and change according to different physiological conditions (e.g., during exercise), and therefore, the HF component can sometimes appear outside the standard HF band limits. In the software, the respiratory frequency is estimated from the ECG signal (if available), more precisely from the R-wave amplitudes which are known to change due to respiration related chest movements [19]. The ECG derived respiration (EDR) is not as accurate as, for example, respiratory flow measurements, but it can be considered to give reliable estimates of the respiratory frequency.

2.4. *Nonlinear methods*

Because of the complex control system of the heart it is presumable that HRV can not be fully described using linear methods. Therefore, various nonlinear methods have been applied to HRV to fully capture the characteristics of the beat-to-beat variability. Nonlinear methods implemented in the software are: Poincaré plot, approximate entropy (ApEn), sample entropy (SampEn), correlation dimension (D_2), detrended fluctuation analysis (DFA), and recurrence plot (RP) analysis.

The Poincaré plot is a graphical presentation of the correlation between consecutive RR intervals, i.e. a plot of RR_{j+1} as a function of RR_j. The shape of the plot is quantified by fitting an ellipse into the data points (RR_j,RR_{j+1}) oriented along the line-of-identity (LOI; line where $RR_j = RR_{j+1}$) [20]. The width and length of the ellipse are determined by the standard

deviations of the points perpendicular to and along the LOI and they are denoted by SD1 and SD2, respectively. SD1 can be considered to measure short-term variability, whereas SD2 measures long-term variability.

The complexity or irregularity of the HRV can be measured by ApEn and SampEn. The computation of these entropy measures depends on two parameters, i.e. the embedding dimension m and the tolerance r (for details see [21,22]). The default values for these parameters are $m = 2$ and $r = 0.2$ SDNN. The tolerance is fixed in relation to SDNN so that these complexity measures are not sensitive to the overall variability level and results of different subjects can be compared.

DFA measures the correlations within the data for different time scales [25,26]. In HRV analysis, these correlations are divided into short-term and long-term fluctuations which are characterized by parameters α_1 and α_2, respectively. These parameters are slopes of a log-log plot (correlation measure as a function of segment length). In the software, the short-term fluctuations slope α_1 is obtained from the plot by default within range of 4–16 beats and α_2 within range of 16–64 beats.

Correlation dimension is another method for measuring the complexity or strangeness of the data. D_2 is expected to give information on the minimum number of dynamic variables needed to model the underlying system. The computation of D_2 depends on the embedding dimension m and threshold r (for details see [23,24]). The default values for these are $m = 10$ and $r = \sqrt{m}$ SDNN.

Yet another approach for analysing the complexity of the data is the so-called recurrence plot analysis. The computation of RP depends on the same embedding dimension (m) and threshold (r) values as D_2. RP is simply a binary square matrix consisting of values 0 and 1; a point in RP gets a value one (1) if the two embedding vectors are close to each others. The RP matrix usually shows short line segments of ones parallel to the main diagonal. The lengths of these lines describe the duration of which the recurring structures are close to each other. Several parameters have been introduced for quantifying these recurrences [27,28]. In the software, the following parameters have been included: mean line length (Lmean), maximum line length (Lmax), recurrence rate (REC), determinism (DET), and Shannon entropy of line length distribution (ShanEn).

3. Software description

Kubios HRV 2.1 has been developed using MATLAB® release 2012a (The MathWorks, Inc.) and was compiled to a deployable standalone application with the MATLAB® Compiler 4.17. The MATLAB® Compiler Runtime (MCR) version 7.17 is required for running Kubios HRV.

3.1. Input data formats

Kubios HRV supports both binary and ASCII text files. The input must be either raw ECG data or beat-to-beat RR interval data. The following file formats are supported:

1. Biopac AcqKnowledge (Biopac Systems Inc.) (*.acq)
2. European data format (EDF) (*.edf)
3. General data format (GDF) (*.gdf)
4. ECG ASCII data files (*.txt,*.dat)
5. Polar HRM files (Polar Electro Ltd.) (*.hrm)
6. Suunto SDF/STE files (Suunto Ltd.) (*.sdf,*.ste)
7. RR interval ASCII files (*.txt,*.dat)
8. Custom ASCII data files (*.txt,*.dat)
9. Kubios HRV Matlab MAT files (*.mat)

The first three file formats (ACQ, EDF and GDF) are binary data formats, of which the GDF format is an excellent general purpose data format for biomedical signals [31]. These binary data files may include several recorded channels from which the software tries to identify the ECG channel automatically based on the channel labels. If the channel can not be identified, the software prompts the user to select the channel. A limited support for multi-channel data in ASCII format is also included, but the data need to be organized in columns. RR interval data is supported in ASCII text file format and in the file formats of two popular heart rate monitor manufactures (Polar Electro Ltd. and Suunto Ltd.).

It should be noted that analysis of long ECG recordings as a whole may not be possible due to computer memory usage. In such a situation, one can select to open shorter episodes (e.g. few hours at a time) of ECG data for analysis. When opening a long ECG recording, the software automatically prompts the user to select if he/she wants to open the recording as a whole or part of the recording.

Finally, the software supports Matlab MAT files saved by Kubios HRV itself. Such a result file includes all the analysis parameters and results, as well as the raw data (ECG or RR data), and therefore, you can easily return to already analysed data by opening the saved MAT file in the software. The MAT files are also useful if you wish to post-process the results in Matlab.

3.2. The user interface

Kubios HRV is fully operated through its graphical user interface which can be divided into four segments as shown in Fig. 1: (1) RR interval series options, (2) Data browser, (3) Analysis options, and (4) Results view segments. The functionalities included in these segments are described below.

3.2.1. RR interval series options

RR interval series options segment shown in Fig. 1 includes artefact correction, sample selection and trend removal options. By using the artefact correction option, artefacts due to ectopic beats, missed beat detections, etc. can be corrected by choosing an appropriate correction level (threshold for detecting artefact beats) which removes the artefacts but does not distort normal RR intervals. The corrections to be made on the RR series are displayed on the RR interval axis. When the corrections are applied, detected artefact beats are replaced using cubic spline interpolation. If ECG is measured, any misdetections should be corrected by editing the R-peak

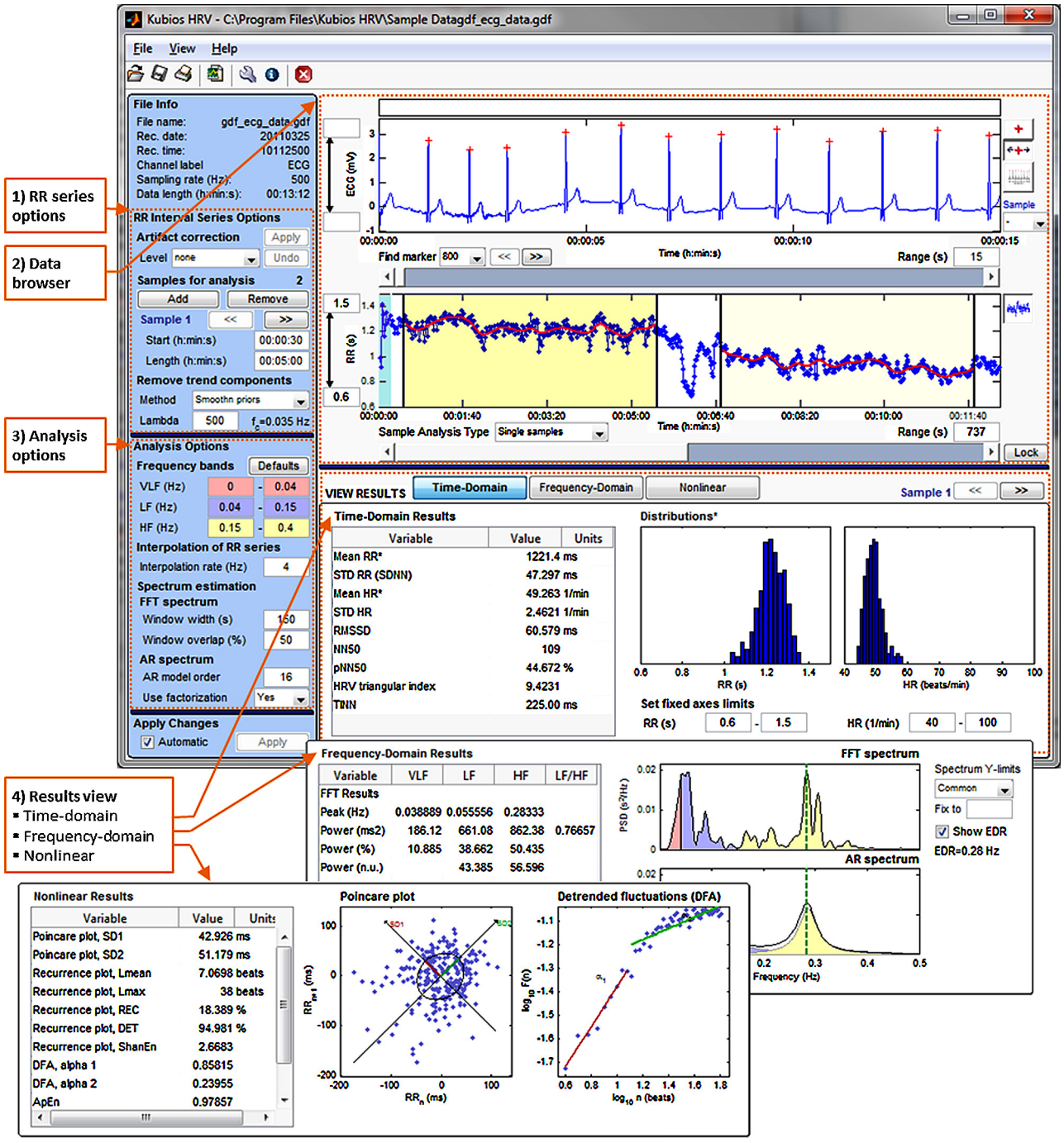

Fig. 1 – The graphical user interface of Kubios HRV, divided into four segments: (1) RR interval series options, (2) data browser, (3) analysis options and (4) results view. Results view is divided into time-domain, frequency-domain and nonlinear results.

detections displayed on the ECG axis as described in Section 3.2.2.

It is possible to select a number of episodes for analysis out of a longer recording. The samples to be analysed can be modified by adding or removing samples and by changing the start time and length of the sample. If more than one sample is selected, the analysis can be done either for the single samples separately or by merging the samples into one longer sample before analysis. This selection is visible under the RR series axis when multiple samples are selected. The starting point and length of the samples can also be changed by moving/resizing the patch over the RR series as described in Section 3.2.2. This section also describes how to add or remove samples from the RR series axes.

The remove trend components options can be used for removing any disturbing low frequency trend components from the RR interval series. Detrending can have a remarkable effect on the analysis results, because all analyses (with

only few exceptions mentioned in Section 3.2.4) are performed from detrended data (if detrending is applied). Detrending options include removal of the first, second, or third order linear trend or the trend can be removed using a method called smoothness priors which was presented in [32]. In the smoothness priors method, the smoothness of the removed trend can be adjusted by editing the Lambda value. The smoothness priors method is basically a time-varying high-pass filter and its cut-off frequency can be adjusted with the Lambda parameter (the bigger the value of Lambda the smoother is the removed trend). The estimated cut-off frequency for the given Lambda value is presented next to the Lambda value edit box. In addition, the trend to be removed from the RRI series is shown over the RR series sample as a red line.

3.2.2. *Data browser*

The data browser segment shown in Fig. 1 displays the measured ECG signal and the extracted RR interval series. If only RR interval data is given as input, the ECG axis will not be displayed. The ECG and RR interval data can be scrolled with the two sliders. The data range shown in the ECG axis is displayed as a green patch over the RR axis. This patch can also be moved with the left mouse button. The range of both axes can be changed by editing the Range values and also the Y-limits of the axes can be manually changed by editing the edit boxes on the left hand side of the axes. The ECG and RR interval axes can also be scrolled together by locking the axes by pressing the Lock button on the right side of the RR axis slider.

In addition to data visualization, the main function of this segment is correction of corrupted RR interval values. If only RR data is available, the artefact correction is performed as described above (Section 3.2.1). However, if ECG is measured, the corrections should be made by editing the misdetected R-peak as follows. Each detected R-peak is marked in the ECG axis with a red "+" mark. Each mark can be moved or removed by right clicking it with the mouse. In addition, new R-peak marks can be added by either right clicking an existing mark and selecting "Add" or by pressing the uppermost button on the right hand side of the ECG axis. Moved or added R-peak marks are by default snapped to local ECG maximum, but manual positioning can also be achieved by pressing the middle button on the right hand side of the ECG axis. The changes made in R-peak marks will be automatically updated to RR interval series.

The RR interval samples selected for analysis are shown as yellow patches on the RR axis and they can be modified with mouse as follows. Each sample can be moved or resized by grabbing it with the left mouse button. You can also add a new sample to a specific location in the RR series by right clicking the RR axis. The new sample will start from the clicked time instant and the length of the sample is by default equal to the length of the previous sample. After right clicking the RR axis a small pop-up window opens in which the sample start time and length can be accepted/modified. When more than one sample exist, a sample can be removed by right clicking it with the mouse.

In addition, the data browser segment includes buttons for displaying a printout of the ECG recording (on the right hand side of the ECG axis), moving the ECG axis view to the beginning of a selected sample (on the right hand side of the ECG axis), scrolling the event markers of the recording session (below the ECG axis), and changing the RR series display mode (on the right hand side of the RR axis).

3.2.3. *Analysis options*

The analysis options segment shown in Fig. 1 includes only HRV frequency-domain analysis options such as HRV frequency band settings, interpolation of RR series, and spectrum estimation options. Other analysis options can be adjusted by editing software preferences as described in Section 3.4. VLF, LF and HF frequency bands can be adjusted by editing the corresponding edit boxes. The default values for these bands are VLF: 0–0.04 Hz, LF: 0.04–0.15 Hz, and HF: 0.15–0.4 Hz according to [5]. These default values can be restored by pressing the "Defaults" button.

The RR interval time series is an irregularly sampled series, and thus, the RR series is converted into equidistantly sampled form by using cubic spline interpolation prior to spectrum estimation. The sampling rate of the interpolation can be adjusted and the default value of it is 4 Hz.

Under the spectrum estimation options, the window width and window overlap values related to Welch's periodogram method can be adjusted. The default values of these are 256 s and 50%, respectively. Related to AR spectrum, the order of the AR model can be adjusted (default value is 16) and the user can select whether or not to use spectral factorization. Note, that the spectral factorization somewhat changes the band power values, because they are computed differently (see Section 2.3).

3.2.4. *Results view*

The results for the selected RR interval sample are displayed in the results view segment shown in Fig. 1. The results are divided into time-domain, frequency-domain and nonlinear results. The results of each section are displayed by pressing the corresponding button on the top of the results view segment. The results are by default updated automatically whenever any one of the sample or analysis options that effect on the results is changed. The updating of the results can be time consuming for longer data samples, in which case it might be useful to disable the automatic update by unchecking the "Automatic" check box in the bottom left corner of the user interface.

The time-domain results view displays the time-domain variables in a table and the RR interval and heart rate histograms in the two axes. Most of the results are calculated from the detrended RR series (if detrending is applied), but there are two obvious exceptions (i.e. mean RR interval and mean HR) which are marked with the $*$ symbol.

The frequency-domain results view displays the results for both FFT and AR spectrum estimation methods. The spectra of the two methods are presented in the two axes. The Y-limits of the two spectra (i.e. the power scale) can be adjusted with the options on the upper right corner of the frequency-domain results view. The power scale can be either common or distinct for the two spectra. If common Y-limits are selected, the limit can be entered manually into the edit box below the selection button. The selected power axis options apply also for the report sheet. The results for both spectra are displayed in the table beside the axes. If ECG is measured, an estimate of

the respiratory frequency, i.e. the EDR, is also computed and shown as a vertical line in both spectrum estimates.

The nonlinear results view displays all the calculated nonlinear variables in one table. In addition, the Poincaré plot and the DFA results are also presented graphically in the two axes. In the Poincaré plot, the successive RR intervals are plotted as blue dots and the SD1 and SD2 variables obtained from the ellipse fitting technique are illustrated as red and green lines. In the DFA plot, the detrended fluctuations $F(n)$ are presented as a function of n in a log-log scale and the slopes for the short term and long term fluctuations α_1 and α_2, respectively, are indicated.

3.3. *Saving the results*

There are three different formats in which the results can be saved, i.e. the results can be saved in an ASCII text file, a report sheet of results can be saved in a PDF-file, or the results can be saved in a Matlab MAT-file.

By default the saved ASCII text file includes software and user information, used analysis parameters, and all the analysis results; all in a comprehensible layout. The columns of the text file are separated with semicolons, and thus, the file can be imported into spreadsheet programs such as the Microsoft Excel® for further inspection. Alternatively, the analysis results can be saved in an "SPSS friendly" format (which need to be selected from software preferences), in which case all the parameter values are saved in one row with column headers indicating the corresponding parameter names. If more than one sample have been analysed, the results for different samples are presented in separate rows. The "SPSS friendly" save option is probably the easiest way to produce a single results table, e.g. for performing group level statistics.

The report sheet shown in Fig. 2 presents all the analysis results in a compact printable form. The report sheet can be generated by selecting "Report sheet" from the View menu or by pressing the corresponding toolbar button. If multiple samples have been selected for analysis, a report sheet is generated for each sample. The generated report sheet(s) can be exported to PDF or printed. When Save Results have been selected, the report sheet(s) can be saved in a single PDF-file by selecting Report figure as the saving type in the save dialog. In this case, the report sheet(s) will not be displayed, but just saved in the selected PDF-file.

The analysis results can also be saved in a Matlab MAT-file (compatible with Matlab version 5 or higher). The MAT-file includes a single structured array variable named `Res` which includes under the field `HRV` all the analysis results, used analysis parameters and the RR data. In addition, the name and path of the original data file as well as the original raw data are saved in the `Res` structure. This save option is useful for users who want to further analyse or process the HRV data in Matlab.

3.4. *Software preferences*

The software includes several analysis options, the preference values of which are designed for short-term human HRV recordings. Some of these analysis options can be adjusted in the user interface, but changes in user interface apply only for the current session. In order to change the preference values, select Edit Preferences from the File menu or press the corresponding toolbar button. This will open the preferences window shown in Fig. 3. The preferences are divided into four categories: user information, analysis options, advanced settings, and report settings. The user information (name, department and organization) entered into preferences is shown on the report sheet and is included in saved result files. Under the analysis options (shown in Fig. 3), the default input data type, number of samples to be analysed, detrending method and frequency band limits can be adjusted. Under the advanced settings, the defaults for spectrum estimation options as well as certain nonlinear analysis options (which are not adjustable from the user interface), can be adjusted. Finally, under the report settings, the paper size of the report sheet can be changed (A4 or letter paper), specifications of the saved ASCII text file can be adjusted (field delimiter, decimal point and the "SPSS friendly" format), and a custom print command can be specified. Changes to the preferences are saved by pressing the "OK" button and they come into effect in the next analysis session.

In addition to the above mentioned preferences, there are various other editable options within the user interface which have mainly influence on the usability of the software. Such options include the Range and Y-limit values of the data axes and various visualization options. The values of these options are preserved in memory and any changes made to them will be applied in the future sessions.

4. Sample run

As a sample run, HRV analysis for the lying and standing periods of the orthostatic test was performed. The sample data (GDF-file `gdf_ecg_data.gdf`, distributed with the software) was measured from a healthy young male subject. During the orthostatic test, a strong increase in heart rate is observed immediately after standing up, which aims to compensate the sudden decrease in blood pressure. In addition, a strong decrease in HF component power and a more gradual increase in LF power are typically observed when subject stands up. The lying and standing periods of the orthostatic test were analysed within one session as follows.

Once the data file was opened in the user interface, the samples to be analysed were selected. By default, the whole recording is selected for analysis. In order to analyse the lying period, the length of the analysis sample was fixed to 5 min and the sample was centred at the lying period. A new analysis sample was then produced by pressing the "Add" button and the new sample was centred at the standing period (the length of the new sample was equal to the first one by default). The two selected samples are shown in Fig. 1. Since we wanted to analyse the two samples separately and compare the results, we then checked that the sample analysis type option under the RR axis was set for "Single samples".

Next, we edited different analysis options to fit our purposes (if we were to analyse several data files with the same settings, the preference values could have been changed). Now we were interested in changes in the LF and HF components, and thus, we removed the very low frequency trend

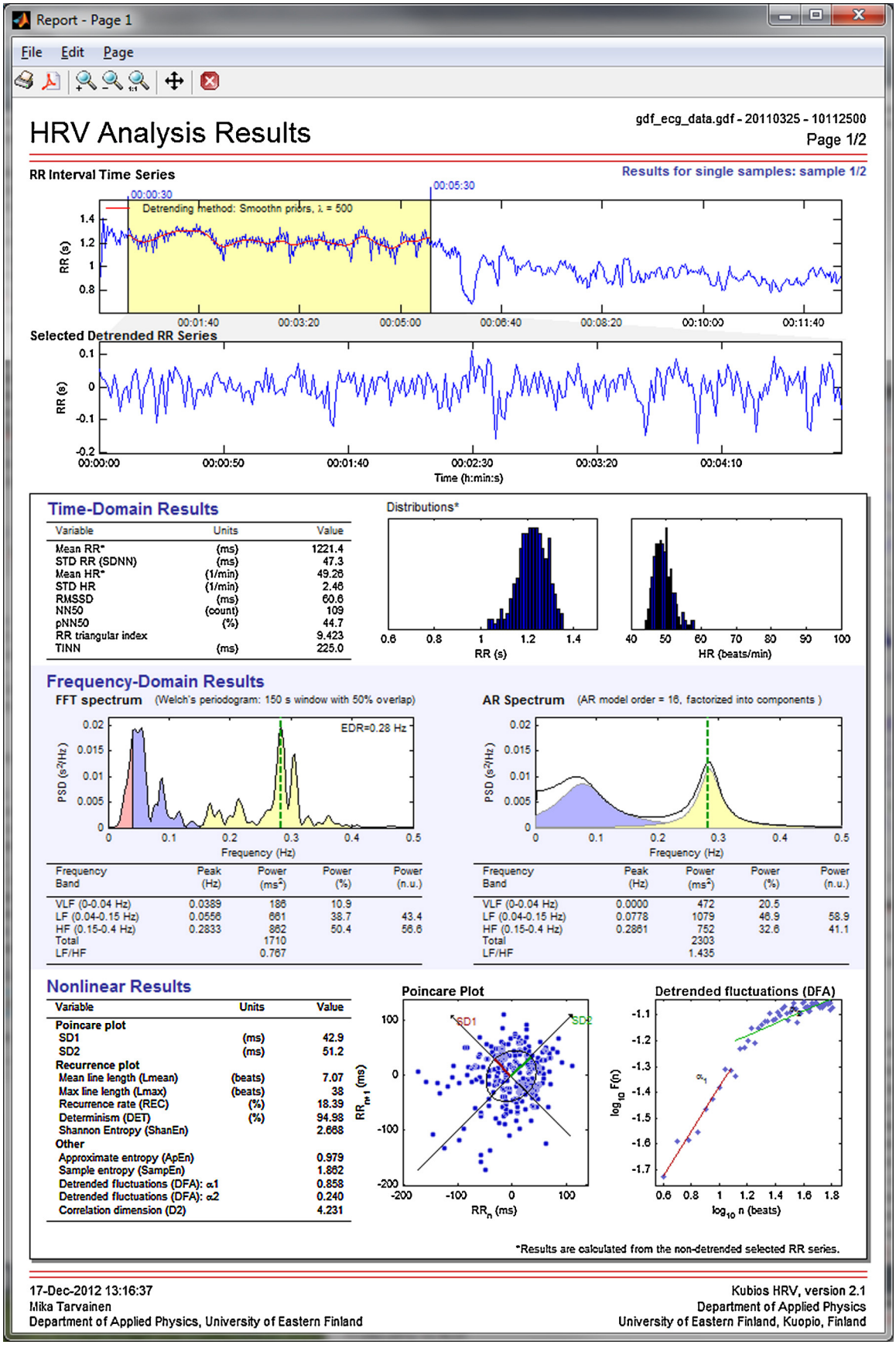

Fig. 2 – The report sheet of Kubios HRV.

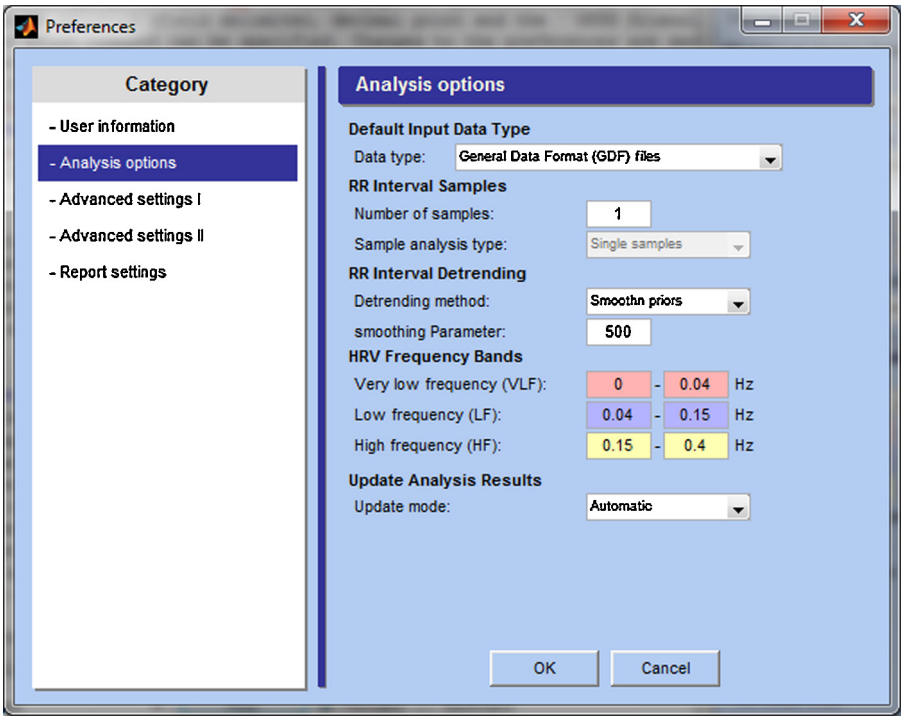

Fig. 3 – Preferences window of Kubios HRV.

components by using the smoothness priors method. The smoothing parameter was set to $\lambda = 500$ which corresponds to a cut-off frequency of 0.035 Hz (below LF frequency band). In addition, for Welch's periodogram (the "FFT spectrum") we set the window width to 150 s and overlap to 50% and in AR spectrum we selected to use spectral factorisation. Otherwise default values were used in the analysis.

The time-domain, frequency-domain and nonlinear analysis results for the two samples could then be viewed in the results view segment of the user interface as shown in Fig. 1 (showing results for the first sample). The results for the second sample could be viewed by pressing the "≫" button on the top right corner of the results view segment (the text on the left indicates which sample's results are shown, the sample is also highlighted in the RR series axis).

Finally, the analysis results were saved in different formats. This was done by selecting "Save Results" from the File menu and then `Save all (*.txt,*.mat,*.pdf)` as the save type and by entering a file name (without any extension). The numeric results of the analysis were saved in the `*.txt` text file and in the `*.mat` MATLAB file, and the report sheets in the `*.pdf` file. The generated PDF-file included two pages (first page showing results for the lying period and second for the standing period). The first page of the PDF-file is a printout of the report sheet shown in Fig. 2 (without the figure window's menubar and toolbar). In the text file, the results for the two samples are presented side by side as can be seen from Fig. 4 (a). Note that you can also save the results in a "SPSS friendly" format (this option can be selected from the software preferences) in which case the output text file would appear as shown in Fig. 4 (b).

5. Availability of the software

Kubios HRV 2.1 (for Windows and Linux operating systems) is available free of charge for non-commercial use for researchers, clinicians, fitness enthusiasts, etc. at http://kubios.uef.fi. For support on using Kubios HRV please see the User's Guide and FAQ (available at http://kubios.uef.fi) or email us at kubios-software@uef.fi.

6. Discussion

In this paper, the Kubios HRV software (ver. 2.1) was introduced. The software includes a wide variety of time-domain, frequency-domain and nonlinear HRV parameters. The software supports several ECG and RR interval data formats and performs the necessary pre-processing steps (QRS detection, artefact correction, and detrending). In addition, ECG derived respiration is included to provide an estimate of the respiratory frequency. Different pre-processing and analysis options can be modified by the user. The software is fully operated through an easy to use graphical user interface.

The standard time and frequency-domain measures of HRV are computed according to the guidelines given in [5] which is important to attain good comparability between the results of HRV studies carried out in different branches of science. Thereby, it is advisable to include these standard parameters (or at least some of them) as reference values for any study.

Few points which can have a significant effect on the analysis results need to be emphasized here. Firstly, any artefacts in the RR interval series should be carefully corrected before

(a)

gdf_ecg_data_hrv.txt - Notepad

File Edit Format View Help

```
RR Interval Samples Selected for Analysis
                          ;    Sample 1;    Sample 2;
  Sample limits (s):      ;      30-330;     407-707;
Sample Analysis Type: Single samples


RESULTS FOR SINGLE SAMPLES
                          ;    SAMPLE 1;           ;    SAMPLE 2;           ;
Time-Domain Results       ;
 Statistical parameters   ;
  Mean RR  (ms):          ;  1221.3908;           ;    928.2578;           ;
  STD RR (ms):            ;    47.2970;           ;     39.1585;           ;
  Mean HR (1/min):        ;    49.2631;           ;     64.9545;           ;
  STD HR (1/min):         ;     2.4621;           ;      3.3990;           ;
  RMSSD (ms):             ;    60.5794;           ;     26.2856;           ;
  NN50 (count):           ;        109;           ;          15;           ;
  pNN50 (%):              ;    44.6721;           ;      4.6584;           ;
  SDANN (ms):             ;           ;           ;           ;           ;
  SDNN index (ms):        ;           ;           ;           ;           ;
 Geometric parameters     ;
  RR tri index:           ;    9.423077;           ;    9.500000;           ;
  TINN (ms):              ;    225.0000;           ;    195.0000;           ;

Frequency-Domain Results  ;FFT spectrum; AR spectrum;FFT spectrum; AR spectrum;
 Peak frequencies         ;
  VLF (Hz):               ;    0.038889;    0.000000;    0.033333;    0.000000;
  LF (Hz):                ;    0.055556;    0.077778;    0.091667;    0.077778;
  HF (Hz):                ;    0.283333;    0.286111;    0.152778;    0.213889;
 Absolute powers          ;
  VLF (ms^2):             ;    186.1189;    472.1776;    386.4708;       0.0000;
  LF (ms^2):              ;    661.0752;   1079.2484;   1056.6440;   1300.7852;
  HF (ms^2):              ;    862.3782;    751.9420;    347.0730;    217.2609;
```

(b)

Microsoft Excel - gdf_ecg_data_hrv_2.txt

	A	B	C	D	E	F	G	H	I	J	K	L
1	Sample	meanrr	sdnn	meanhr	sdhr	rmssd	nn50	pnn50	sdann	sdnni	hrvtri	tinn
2	1	1221.391	47.29696	49.26314	2.462134	60.57938	109	44.67213			9.423077	225
3	2	928.2578	39.15848	64.95447	3.398972	26.28558	15	4.658385			9.5	195
4												

gdf_ecg_data_hrv_2

Ready 100%

Fig. 4 – Results of the sample run saved in the two alternative text file formats (a) the default format and (b) the "SPSS friendly" format.

analysis because even single artefacts can have a significant effect on analysis results. The effect of artefacts is biggest on parameters which reflect HF variability (peaks in data increase power at higher frequencies). Secondly, a trend (i.e. a slow change in mean heart rate during the recording) can have a big impact on HRV parameter values. This concerns mainly HRV parameters reflecting overall variability (e.g. SDNN and TINN), but also the estimation of LF component from AR spectrum can be highly distorted as shown in [32]. Thus, in short-term analysis when only LF and HF components are of interest, the very low frequency trend components should be removed using the detrending options. Finally, the HF component reflecting parasympathetic nervous activity is known to appear at the respiratory frequency, and thus, the respiratory frequency should be incorporated into HRV analysis if possible in order to avoid misinterpretations of the analysis results.

It is clear that all non-commercial HRV software tools, including the ones proposed in [11–16], have their strengths and thereby their users. Compared to other software tools, Kubios HRV is a complete solution for HRV analysis with support for several data formats (both ECG and RR data), built-in preprocessing options (QRS detection, artifact correction and detrending), wide variety of analysis methods (computing all standard time and frequency-domain parameters and several nonlinear parameters) and easy-to-use graphical user interface. Furthermore, Kubios HRV includes certain features which are not available in any other software such as the spectral factorization (decomposition of the AR spectrum into VLF, LF and HF components) and built-in respiratory frequency estimation from the ECG data.

REFERENCES

[1] G. Berntson, J.B. Jr., D. Eckberg, P. Grossman, P. Kaufmann, M. Malik, H. Nagaraja, S. Porges, J. Saul, P. Stone, M.V.D. Molen, Heart rate variability: origins, methods, and interpretive caveats, Psychophysiology 34 (1997) 623–648.

[2] T. Laitio, J. Jalonen, T. Kuusela, H. Scheinin, The role of heart rate variability in risk stratification for adverse postoperative cardiac events, Anesth. Analg. 105 (6) (2007) 1548–1560.

[3] M. Pagani, N. Montano, A. Porta, A. Malliani, F. Abboud, C. Birkett, V. Somers, Relationship between spectral components of cardiovascular variabilities and direct measures of muscle sympathetic nerve activity in humans, Circulation 95 (1997) 1441–1448.

[4] R. Furlan, A. Porta, F. Costa, J. Tank, L. Baker, R. Schiavi, D. Robertson, A. Malliani, R. Mosqueda-Garcia, Oscillatory patterns in sympathetic neural discharge and

cardiovascular variables during orthostatic stimulus, Circulation 101 (2000) 886–892.
[5] Task force of the European society of cardiology and the North American society of pacing and electrophysiology, Heart rate variability – standards of measurement, physiological interpretation, and clinical use, Circulation 93 (5) (1996) 1043–1065.
[6] U. Acharya, K. Joseph, N. Kannathal, C. Lim, J. Suri, Heart rate variability: a review, Med. Biol. Eng. Comput. 44 (2006) 1031–1051.
[7] C. van Ravenswaaij-Arts, L. Kollée, J. Hopman, G. Stoelinga, H. van Geijn, Heart rate variability, Ann. Intern. Med. 118 (6) (1993) 436–447.
[8] M. Malik, A. Camm, Components of heart rate variability – what they really mean and what we really measure, Am. J. Cardiol. 72 (11) (1993) 821–822.
[9] J. Pumprla, K. Howorka, D. Groves, M. Chester, J. Nolan, Functional assessment of heart arte variability: physiological basis and practical applications, Int. J. Cardiol. 84 (2002) 1–14.
[10] J. Achten, A. Jeukendrup, Heart rate monitoring – applications and limitations, Sports Med. 33 (7) (2003) 517–538.
[11] R. Maestri, G. Pinna, POLYAN: a computer program for polyparametric analysis of cardio-respiratory variability signals, Comput. Methods Prog. Biomed. 56 (1998) 37–48.
[12] J. de Carvalho, A. da Rocha, F. de Oliveira Nascimento, J.S.L.J. Neto Jr., Development of a Matlab software for analysis of heart rate variability, in: B. Yuan, X. Tang (Eds.), 6th international conference on signal processing, Institute of electrical and electronics engineering, Beijing, China, 2002, pp. 1488–1492.
[13] P. Perakakis, M. Joffily, M. Taylor, P. Guerra, J. Vila, KARDIA: a Matlab software for the analysis of cardiac interbeat intervals, Comput. Methods Prog. Biomed. 98 (2010) 83–89.
[14] J. Mietus, A. Goldberger, Heart rate variability analysis with the HRV toolkit: basic time and frequency domain measures, Available online: http://www.physionet.org/tutorials/hrv-toolkit/ (accessed 2.9.2013).
[15] L. Rodríguez-Li nares, A. Méndez, M. Lado, D. Olivieri, X. Vila, I. Gómez-Conde, An open source tool for heart rate variability spectral analysis, Comput. Methods Prog. Biomed. 103 (2011) 39–50.
[16] K. Kudryński, P. Strumillo, J. Ruta, Computer software tool for heart rate variability (HRV), T-wave alternans (TWA) and heart rate turbulence (HRT) analysis from ECGs, Med. Sci. Monit. 17 (9) (2011) MT63–71.
[17] J.-P. Niskanen, M.P. Tarvainen, P.O. Ranta-aho, P.A. Karjalainen, Software for advanced HRV analysis, Comput. Methods Prog. Biomed. 76 (1) (2004) 73–81.
[18] M.P. Tarvainen, J.-P. Niskanen, J.A. Lipponen, P.O. Ranta-aho, P.A. Karjalainen, Kubios HRV – a software for advanced heart rate variability analysis, in: ECIFMBE, IFMBE Proc, vol. 22, 2009, pp. 1022–1025.
[19] G. Moody, R. Mark, A. Zoccola, S. Mantero, Derivation of respiratory signals from multi-lead ECGs, Comput. Cardiol. 12 (1985) 113–116.
[20] M. Brennan, M. Palaniswami, P. Kamen, Do existing measures of Poincaré plot geometry reflect nonlinear features of heart rate variability, IEEE Trans. Biomed. Eng. 48 (11) (2001) 1342–1347.
[21] J. Richman, J. Moorman, Physiological time-series analysis using approximate entropy and sample entropy, Am. J. Physiol. 278 (2000) H2039–H2049.
[22] Y. Fusheng, H. Bo, T. Qingyu, Approximate entropy and its application in biosignal analysis, in: M. Akay (Ed.), Nonlinear Biomedical Signal Processing: Dynamic Analysis and Modeling, vol. II, IEEE Press, New York, 2001, pp. 72–91, Ch. 3.
[23] S. Guzzetti, M. Signorini, C. Cogliati, S. Mezzetti, A. Porta, S. Cerutti, A. Malliani, Non-linear dynamics and chaotic indices in heart rate variability of normal subjects and heart-transplanted patients, Cardiovasc. Res. 31 (1996) 441–446.
[24] B. Henry, N. Lovell, F. Camacho, Nonlinear dynamics time series analysis, in: M. Akay (Ed.), Nonlinear Biomedical Signal Processing: Dynamic Analysis and Modeling, vol. II, IEEE Press, New York, 2001, pp. 1–39, Ch. 1.
[25] C.-K. Peng, S. Havlin, H. Stanley, A. Goldberger, Quantification of scaling exponents and crossover phenomena in nonstationary heartbeat time series, Chaos 5 (1995) 82–87.
[26] T. Penzel, J. Kantelhardt, L. Grote, J.-H. Peter, A. Bunde, Comparison of detrended fluctuation analysis and spectral analysis for heart rate variability in sleep and sleep apnea, IEEE Trans. Biomed. Eng. 50 (10) (2003) 1143–1151.
[27] C. Webber Jr., J. Zbilut, Dynamical assessment of physiological systems and states using recurrence plot strategies, J. Appl. Physiol. 76 (1994) 965–973.
[28] J. Zbilut, N. Thomasson, C. Webber, Recurrence quantification analysis as a tool for the nonlinear exploration of nonstationary cardiac signals, Med. Eng. Phys. 24 (2002) 53–60.
[29] J. Pan, W. Tompkins, A real-time QRS detection algorithm, IEEE Trans. Biomed. Eng. 32 (3) (1985) 230–236.
[30] M.P. Tarvainen, S.D. Georgiadis, P.O. Ranta-aho, P.A. Karjalainen, Time-varying analysis of heart rate variability signals with Kalman smoother algorithm, Physiol. Meas. 27 (3) (2006) 225–239.
[31] A. Schlögl, GDF – a general dataformat for biosignals, arXiv:cs/0608052v8 [cs.DL] (June 2011).
[32] M.P. Tarvainen, P.O. Ranta-aho, P.A. Karjalainen, An advanced detrending method with application to HRV analysis, IEEE Trans. Biomed. Eng. 49 (2) (2002) 172–175.