



# **Kubios HRV Software**

# USER'S GUIDE

# **Kubios HRV Standard Kubios HRV Premium**

(version 3.5)

November 3, 2021

Mika P. Tarvainen, Ph.D.
Jukka Lipponen, PhD
Juha-Pekka Niskanen, PhLic
Perttu O. Ranta-aho, MSc

# Contents

1	Overview	4
	1.1 About Kubios HRV	
	1.2 System requirements	
	1.3 Release notes	
	1.4 Installation	
	1.5 Uninstallation	6
2	Supported data formats and devices	7
3	Beat detection and pre-processing	10
	3.1 Beat detection algorithms	
	3.2 Noise segments	
	3.3 Artefact correction algorithms	
	3.4 IBI time series trend removal	12
4	Kubios HRV user interface	13
	4.1 RR interval series options	13
	4.2 Data browser	
	4.3 Results view	
	4.3.1 Results overview	
	4.3.2 Time-domain results view	
	4.3.3 Frequency-domain results view	
	4.3.5 Time-varying results view	
	4.4 Menus and toolbar buttons	
	1.1 Wellas and toolsal sattons	, ,
5	Saving the results	20
	5.1 ASCII text file	
	5.2 PDF report	
	5.3 Matlab MAT-file (available in Premium)	
	5.4 SPSS Menuly Datch life (available in Premium)	22
6	Kubios HRV preferences	24
7	Kubios HRV analysis parameters	30
Α	Appendix: Polar Flow export (available in Premium)	31
В	Appendix: Kubios HRV figures	34
Re	eferences	40

### **Abbrevations**

AC Heart rate acceleration capacity (AC) index ACQ AcgKnowledge data file format (Biopac Inc.)

ANS Autonomic nervous system
ApEn Approximate Entropy

AR Autoregressive (model or process)

ASCII Text file using the ASCII character set, which is the most common format for English-

language text files

CSV Comma separated value (file format)
DC Heart rate deceleration capacity (DC) index

DFA Detrended fluctuation analysis

ECG Electrocardiogram

EDF European data format (file format)

EE Energy expenditure FFT Fast Fourier transform

GDF General data format (file format)

GUI Graphical user interface

HF High frequency (refers to HRV frequency band, by default 0.15-0.4 Hz)

HR Heart rate

HRV Heart rate variability

IBI Inter-beat-interval (same as RR interval)

LF Low frequency (refers to HRV frequency band, by default 0.04-0.15 Hz)

MAT MATLAB data file format (Mathworks Inc.)

MSE Multiscale entropy

NNxx Number of successive RR interval pairs that differ more than xx msec

PDF Portable document format (file format)

pNNxx Relative number of successive RR interval pairs that differ more than xx msec

PNS Parasympathetic nervous system

PPG Photoplethysmogram (measurement of blood volume changes)

QRS QRS complex of electrocardiogram

RESP Respiration rate

RMSSD Root mean square of successive RR interval differences

RPA Recurrence plot analysis

RR Time interval between successive ECG R-waves (RR interval, same as IBI)

SampEn Sample entropy ShanEn Shannon entropy

SDANN Standard deviation of the averages of RR intervals in 5-min segments

SDNN Standard deviation of normal-to-normal RR intervals

SDNNI Mean of the standard deviations of RR intervals in 5-min segments

SI Stress index

SNS Sympathetic nervous system

SPSS Statistical analysis software package(IBM Corp.)
TINN Triangular interpolation of normal-to-normal intervals

TRIMP Training impulse

VLF Very low frequency (refers to HRV frequency band, by default 0-0.04 Hz)

#### 1 Overview

#### 1.1 About Kubios HRV

Kubios HRV is a gold-standard heart rate variability (HRV) analysis software designed for research and professional use. Kubios HRV software is used at roughly 1200 universities in 128 countries. With Kubios HRV, you can turn your ECG or HR monitor into a powerful tool to probe the cardiovascular system or to evaluate the effect of stress and recovery on heart health. The software is suitable for clinical and public health researchers, professionals working on human well-being, or sports enthusiasts; for anybody who want to perform detailed analyses on heart rate variability, e.g. to examine autonomic nervous system function.

The first versions of the Kubios HRV were developed as part of academic research work carried out at the Department of Applied Physics, University of Eastern Finland, Kuopio, Finland. Earlier versions of Kubios HRV has been described in two scientific publications [19, 28]:

Niskanen J-P, Tarvainen MP, Ranta-aho PO, and Karjalainen PA. Software for advanced HRV analysis. *Comp Meth Programs Biomed*, 76(1):73-81, 2004.

Tarvainen MP, Niskanen J-P, Lipponen JA, Ranta-aho PO, and Karjalainen PA. Kubios HRV – Heart rate variability analysis software. *Comp Meth Programs Biomed*, 113(1):210-220, 2014.

Kubios HRV is a scientifically validated software and the most commonly used HRV analysis software for scientific research. This is evidenced by the fact that the two publications listed above have attracted over 1 000 citations.

Kubios HRV is currently available as two alternative products:

**Kubios HRV Standard:** Freeware HRV analysis software for non-commercial personal use. Supports HR data from most common HR monitor manufacturers and computes most commonly used time- and frequency-domain HRV parameters. Software is operated through an easy-to-use GUI and analysis results can be saved as PDF report or text file.

Product page: https://www.kubios.com/hrv-standard.

**Kubios HRV Premium:** Full featured HRV analysis software designed for scientific research and professional use. Supports wide range of ECG, PPG and HR data and computes all commonly used time-domain, frequency-domain and nonlinear HRV parameters. In addition, Kubios HRV Premium includes improved preprocessing (beat correction and noise handling), ECG derived respiration, time-varying analysis and extended exporting options. Analysis results can be saved as illustrative PDF reports, CSV text file, MATLAB MAT file and also in a "SPSS friendly" batch file.

**Product page:** https://www.kubios.com/hrv-premium.

A summary of the features is given in Table 1.

Please visit at <a href="https://www.kubios.com/">https://www.kubios.com/</a>, where you can find current information on the software, download updates, find tutorials and information about HRV and how to use the software. If you have any trouble or questions regarding the software, please check first if your particular problem or question has an answer in the FAQ/troubleshooting section at the software homepage! You can also follow us on Facebook, LinkedIn, Twitter, Instagram and YouTube.

#### 1.2 System requirements

Kubios HRV was developed using MATLAB<sup>®1</sup> and was compiled to a standalone application with the Matlab Compiler. Therefore, in addition to Kubios HRV the MATLAB Runtime also needs to be installed. The latest installers for Kubios HRV and MATLAB Runtime can be downloaded from <a href="https://www.kubios.com/download">https://www.kubios.com/download</a>. Please note that the MATLAB Runtime is free and does not require the user to have a Matlab license. System requirements for running Kubios HRV are similar to those of Matlab (see <a href="https://www.mathworks.com/support/sysreq/">https://www.mathworks.com/support/sysreq/</a>) and only 64-bit operating systems are supported. Please note that **correct version of MATLAB Runtime (available at Kubios download page) must be installed** in order to run Kubios HRV.

<sup>&</sup>lt;sup>1</sup>MATLAB<sup>®</sup>, The Mathworks, Inc.

1.2 System requirements

Table 1: Summary of Kubios HRV Standard and Kubios HRV Premium features

Data support  ■ IBI or RR interval data files: Garmin and Suunto FIT files, Polar TXT files, custom		
• IBI or RR interval data files: Garmin and Suunto FIT files, Polar TXT files, custom		
formatted text and CSV files	<b>✓</b>	<b>✓</b>
ECG/PPG data files: EDF/EDF+, GDF, Biopac ACQ3, Cardiology XML, ISHNE Holter ECG, Physionet MIT, and custom formatted text and CSV files	_	<b>✓</b>
Supported HR monitors: ActiHeart, emWave, Firstbeat Bodyguard, Garmin (Forerunner and Fenix series), Polar (V800), Suunto (Ambit and Spartan series), Zephyr BioHarness	<b>✓</b>	~
+ Direct export from Polar Flow (RR/IBI data from all Polar Flow compatible monitors when H6, H7 or H10 heart rate sensor is used in measurement)	_	✓
Supported ECG/PPG devices: Actiwave Cardio, AliveCor Kardia, Biopac, Bittium Faros, Empatica E4, Mindfield MindMaster, Shimmer and several clinical Holter and ECG monitors	_	✓
Pre-processing		
Built-in QRS detector for accurate detection of ECG R-waves and pulse wave detector for PPG data	_	✓
Automatic noise detection (automatically identifies noisy ECG/RR data segments, manual editing also possible)	_	<b>✓</b>
Beat correction methods: ECG based beat detection corrections / Automatic beat correction / Threshold based beat correction	-/-/ <b>~</b>	<b>\</b> /\ <b>/</b> /\
• Smoothness priors method for removing very low frequency trend components when performing short-term HRV analysis	✓	<b>✓</b>
Analysis options		
Automatic analysis sample generation (based on predefined CSV file)	_	<b>✓</b>
Stress index, PNS index and SNS index	$\checkmark$	<b>✓</b>
Time-domain parameters: Mean RR and HR, min/max HR, SDNN, RMSSD, pNN50, HRV triangular index, TINN etc.	<b>✓</b>	<b>✓</b>
+ HR deceleration (DC) and acceleration capacity (AC)	-	<b>~</b>
Frequency-domain parameters: VLF, LF and HF band powers (in absolute, relative and normalised units), peak frequencies and LF/HF ratio	<b>V</b>	<b>V</b>
Spectrum estimation methods: Welch's periodogram / Lomb-Scargle periodogram / AR spectrum estimate	<b>✓</b> /-/ <b>✓</b>	<b>✓</b> / <b>✓</b> / <b>✓</b>
Basic nonlinear parameters: Poincaré plot, approximate entropy (ApEn), sample entropy (SampEn) and detrended fluctuation analysis (DFA)	<b>✓</b>	<b>✓</b>
$+$ Correlation dimension $(D_2)$ , recurrence plot analysis (RPA), multiscale entropy (MSE)	_	<b>✓</b>
Validated algorithm for respiration rate (RESP) estimation from ECG and RR interval data (useful in reliable RSA component estimation)	-	<b>✓</b>
Time-varying analysis: instantaneous values for energy expenditure (EE), training impulse (TRIMP), stress index, PNS/SNS indexes, and all time-domain and frequency-domain HRV parameters and certain nonlinear parameters. Spectrogram with Fire-colormap	_	<b>✓</b>
Reports and results export		
HRV reports (PDF reports) including: time-domain, frequency-domain and non-linear results / Time-varying analysis results	✓/-	<b>✓</b> / <b>✓</b>
ECG print (PDF report) showing the raw ECG trace for selected time period     HRV analysis results export options: PDF file / CSV text file / MATLAB MAT file	_ <b>~</b> / <b>~</b> / _	414 41414
"SPSS friendly" batch file export (ideal for group analyses or repeated measurements)	_	<b>~</b>

**Windows (7 SP1 or 10, 64-bit)** operating system with 4 GB of RAM, 3-5 GB of disk space, screen resolution of  $1024 \times 768$  or higher, and the MATLAB Runtime installation.

**Mac OS X (High Sierra, Mojave or Catalina)** operating system with 4 GB of RAM, 3-5 GB of disk space, screen resolution of  $1024 \times 768$  or higher, and the MATLAB Runtime installation

**Linux** distribution (see qualified distribution at Mathworks site) with 4 GB of RAM, Intel or AMD x86-64 processor, 4-6 GB of disk space, screen resolution of  $1024 \times 768$  or higher, hardware accelerated graphics card supporting OpenGL 3.3 with 1GB GPU memory, graphical desktop environment, and the MATLAB Runtime installation.

1.3 Release notes 6

#### 1.3 Release notes

The version history of the most significant updates and changes made in Kubios HRV is given in the release notes. Going though the latest release notes is recommended always before updating to a new version. Up-to-date and detailed release notes can be found from <a href="https://www.kubios.com/release-notes">https://www.kubios.com/release-notes</a>.

#### 1.4 Installation

In order to run Kubios HRV, you need to install Kubios HRV and MATLAB Runtime on your computer. Installers can be download from https://www.kubios.com/download. The first time you launch Kubios HRV, you will be prompted to activate the software for the the current computer using your personal License key. In order to get the License key, fill in the order form at https://www.kubios.com/. A short description of the installation process at different operating systems is given below.

**Windows:** Make sure that you have administrative privileges (you will need them to install Kubios HRV). In order to install Kubios HRV on a Windows computer, you need to download and run the Kubios HRV installer. This will install both Kubios HRV and Matlab Runtime on your computer. You can launch Kubios HRV by using the Desktop icon (if created) or by selecting it from the Start Menu. Please note that Kubios HRV also starts the MATLAB Runtime and may take some time especially with older computers. The first time you start Kubios HRV, you also need to activate the software using your personal license key.

**Mac OS:** Download and run the Kubios HRV installer for macOS (available at our download page). The installer will install Kubios HRV and the Matlab Runtime (NOTE: Matlab Runtime will be downloaded during the installation process only if it is not found from your computer) into separate folders under your Applications. To start Kubios HRV, you need to click the Kubios HRV application under the KubiosHRV folder in your Applications. You can also find Kubios HRV application with Spotlight (click on the upper-right corner of the menu bar or press Command+Space). NOTE: If you have an earlier version of Kubios HRV software installed in your Mac, we recommend that you uninstall the older version first (move the installed Kubios HRV and earlier version of Matlab Runtime to trash).

**Linux:** Install the MATLAB Runtime by extracting the MATLAB Runtime zip package and executing the installer command ./install as root. Do not change the default MATLAB Runtime install directory if you do not have a special need to change it. Kubios HRV assumes that MATLAB Runtime is installed in the default directory. Otherwise the MATLAB Runtime directory has to be given as an argument to the run\_kubioshrv script. Kubios HRV can be installed using the deb (Ubuntu/Debian) or rpm (Fedora/SUSE/RedHat) package using your package manager. Furthermore, Kubios HRV can also be installed without a package manager by extracting the tar.gz file to a directory of your choosing. To run Kubios HRV, select it from the menu of your desktop environment or run the command kubioshrv-standard or kubioshrv-premium in the terminal. If you have installed Kubios HRV using the tar.gz package, go to the directory you extracted the package and run ./run\_kubioshrv in the terminal.

#### 1.5 Uninstallation

**Windows:** The software can be uninstalled using the "Windows Settings > System > Apps & Features" (Windows 10) or "Control Panel > Programs and Features" (Windows 7 SP1). However, the uninstaller does not remove your preferences settings or license file. These have to be deleted manually and can be found in the following folders:

#### **Kubios HRV Standard**

- C:\Users\<username>\AppData\Roaming\Kubios\KubiosHRVStandard
- ${\tt C:\ProgramData\Kubios\KubiosHRVStandard}$

#### **Kubios HRV Premium**

- C:\Users\<username>\AppData\Roaming\Kubios\KubiosHRVPremium
- C:\ProgramData\Kubios\KubiosHRVPremium

Mac OS: Move the installed applications (MATLAB Runtime and Kubios HRV application) to trash.

**Linux:** Remove the MATLAB Runtime by deleting the directory it was installed in (default /usr/local/MATLAB/MATLAB\_Runtime by deleting the directory it was installed in (default /usr/local/MATLAB/MATLAB\_Runtime by deleting the directory it was extracted in. However, the preferences and license file have to be deleted manually from the ~/.kubios directory.

## 2 Supported data formats and devices

The different file formats and devices that are compatible with Kubios HRV are described here. Features available only in the Premium version are indicated in the text and an overview of differences between these versions is given in Table 1.

Kubios HRV supports the following data formats:

1. 2. 3.	Polar TXT and HRM files (Polar Electro Ltd.) Suunto FIT files (Suunto Ltd.) Suunto SDF, STE and XML files (older formats) Garmin FIT files (Garmin Ltd.) RR interval text files	(*.txt,*.hrm) (*.fit) (*.sdf,*.ste,*.xml) (*.fit)
4.		(*.txt,*.dat,*.csv)
5.	Custom text data files (only RR data)	(*.txt,*.dat,*.csv)
Avai 6. 7. 8. 9. 10. 11. 12. 13. 14.	, , , , , , , , , , , , , , , , , , , ,	(*.fit) (*.txt,*.dat,*.csv) (*.txt,*.dat,*.csv) (*.acq) (*.xml) (*.edf) (*.gdf) (*.ecg) (*.hea, *.dat) (*.csv)
16.	Kubios HRV Matlab MAT files	(*.mat)

When using Polar, Suunto or Garmin HR monitors, **make sure that your device can measure and store RR intervals in beat-to-beat**. From averaged data (e.g. HR values at every 5 seconds) you are not able to analyse HRV. In addition to standardized HR monitor file formats, a support for plain RR interval text files (ASCII files) is provided. The input text file can include RR interval values in one or two column format, i.e. the RR interval values can be given as:

Type 1	Тур	e 2
0.759	0.759	0.759
0.690	1.449	0.690
0.702	2.151	0.702
0.712	2.863	0.712
0.773	3.636	0.773
:	:	:

In Type 2 format, the first column includes the time indexes of R wave detections (zero time for the first detection in this example) and second column the RR interval values. The RR interval values above are given above in seconds, but also millisecond values can be used.

Similarly, ECG data can be given as input in text file formatted as shown below

Type 1	Ty	pe 2
-0.173	0	-0.173
-0.119	0.002	-0.119
-0.025	0.004	-0.025
0.091	0.006	0.091
0.218	0.008	0.218
:	:	:

where the first column on Type 2 format is the time scale in seconds for the ECG data. The sampling rate of this example file is thus 500 Hz (samples per second). If ECG data is given according to the Type 1 format, user is prompted to enter the sampling rate manually.

In addition to above text file formats, a custom text file option is also provided. Using this option you can import text files including header lines and/or several data columns. Once you have selected an input file, an interface for importing the file into Kubios HRV is opened. This interface and the options that you need to set according to your data file are shown in Fig. 1.

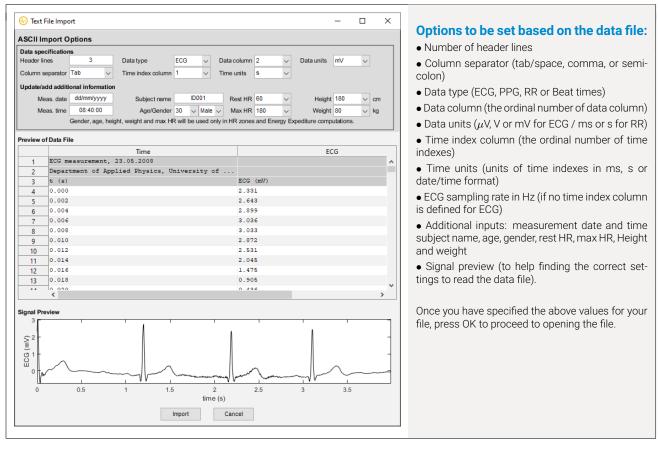


Figure 1: The interface for importing custom text data files into Kubios HRV.

In addition to above text file formats, Kubios HRV supports the Zephyr BioHarness file format (CSV file), Cardiology XML, and five binary data formats (Biopac ACQ3, EDF, GDF, ISHNE ECG, adn Physionet MIT). The EDF/EDF+ are open file formats quite generally used for storing biomedical signal data. The ISHNE ECG is a standard output format for Holter ECG data described in [1]. When any of these binary files are read to the software, Kubios HRV automatically tries to determine the ECG channel from the channel labels. If the ECG channel cannot be determined (or more than one channels are identified as ECG channels), the software prompts the user to select the appropriate channel.

A list of commonly used devices, which are known to be compatible with Kubios HRV is given in Table 2. Please note that this is not a complete list, many other devices may also support exporting the data in Kubios HRV compatible format.

Finally, the software supports also MATLAB MAT files saved by Kubios HRV. When you are using Kubios HRV, you

can save the analysis session into a MATLAB MAT file as described in Section 5.3. The MAT files include all the analysis results and settings as well as the raw data (ECG, PPG or RR data). Therefore, you are able to reopen the analysis session by opening the saved MAT file again in Kubios HRV.

Table 2: Commonly used ECG or HR measurement devices known to be compatible with Kubios HRV.

Devices	Data type	Kubios HRV input option
Actiheart (CamNtech Ltd.)	IBI	RR text file
Actiwave Cardio (CamNtech Ltd.)	ECG	EDF
Biopac system with ECG module (Biopac Systems Inc.)	ECG	Biopac ACQ3
Cortium C3** (Cortrium Ltd.)	ECG	EDF
Faros (Bittium)	ECG/IBI	EDF
Empatica E4 (Empatica Inc.)	PPG	Custom text file
emWave (HeartMath Inc.)	IBI	Custom text file
Firstbeat Bodyguard (Firstbeat Technologies Ltd.)	IBI	SDF file
Garmin (Forerunner and Fenix) HR monitors* (Garmin Ltd.)	IBI	FIT file
GE CardioSoft and CASE systems (GE Healthcare)	ECG	Cardiology XML
Kardia** (AliveCor Inc.)	ECG	EDF
Mindfield MindMaster (Mindfield Biosystems Ltd.)	ECG	EDF
Polar HR monitors* (Polar Electro Ltd.)	IBI	FIT file or HRM/RR text file
Shimmer ECG and PPG systems (Shimmer Sensing)	ECG/PPG	Custom text file
Suunto (Ambit and Spartan) HR monitors* (Suunto Ltd.)	IBI	FIT/SDF/STE/XML files
Zephyr BioHarness (Zephyr Tech. Corp.)	ECG/IBI	Zephyr BioHarness CSV

<sup>\*</sup> Make sure that the model supports beat-to-beat data export (IBI data). A direct export from Polar Flow is available in Premium version (supports any Polar Flow compatible monitor with H6, H7 or H10 heart rate sensor).

<sup>\*\*</sup> Requires converter available from device manufacturer

## 3 Beat detection and pre-processing

#### 3.1 Beat detection algorithms

Kubios HRV Premium has a built-in accurate QRS and pulse wave detectors for detecting heart beats from ECG and PPG data, respectively.

In case ECG data is imported into Kubios HRV Premium, the R-wave time instants are automatically detected by applying the built-in QRS detection algorithm. This in-house developed detection algorithm is based on the Pan-Tompkins algorithm [22]. 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 comparison to expected value between adjacent R-waves. Both of these rules are adjusted adaptively every time a new R-wave is acceptably detected. Before R-wave time instant extraction, the R-wave is interpolated at 2000 Hz to improve the time resolution of the detection. This up-sampling will significantly improve the time resolution of R-wave detection when the sampling rate of the ECG is relatively low (200 Hz or higher recommended).

The pulse wave detector is based on matched filtering approach. Firstly maximum of 1st derivative representing the steepest part of the pulse wave is used for initial pulse location estimation. Secondly, template for the pulse wave (and matched filter) is constructed using the initial pulses. Decision of the final pulse wave locations are defined by comparing the filtered signal against varying threshold and comparing normalized error between the template and PPG signal. Allowed normalized error between template and pulse wave under inspection can be adjusted in software preferences. That is, the smaller the acceptance threshold percent is the more similar the pulse wave have to be with the template in order to be accepted.

The accuracy of the pulse wave detection algorithm is shown in Fig 2. The left panel showing the Bland-Altman plot illustrating the agreement between detected PP intervals and corresponding RR intervals during a resting measurement. The right panel shows error percentages of commonly used HRV parameters estimated from PP interval compared to RR interval time series. Used dataset contains 20 healthy volunteers with wide age scale (20 to 50 years). Error between the RR and PP time series is -0.01 $\pm$ 5.16 ms (mean  $\pm$  SD). This  $\pm$ 5 ms error in heart beat detection produces approximately  $\pm$ 10% maximum errors to the HRV parameters.

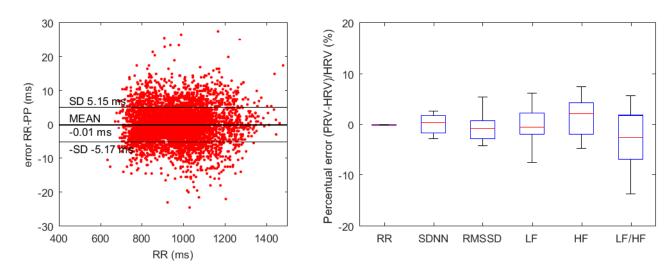


Figure 2: Accuracy of the PP interval vs. RR interval during resting measurement is presented on left panel. In right panel, errors between the PRV and HRV parameters are presented. Blue box indicates region between the 25-75 percentile and black lines are maximum and minimum value.

#### 3.2 Noise segments

Kubios HRV Premium software supports automatic noise detection, which automatically identifies noise segments based on the raw ECG data (if available) and from the interbeat interval data (RR or pulse-to-pulse intervals). The

noise detection level can be changed between Very low, Low, Medium and Strong (default Medium) and you can turn it off by switching it to None "level". Automatic noise detection is extremely helpful for those analyzing long-term ambulatory ECG or RR recordings, where signal quality is not good throughout the recording. Please note that noise detection aims to identify noise segments which distort several consecutive beat detections, and thus, would affect on HRV analysis accuracy. Individual intermittent abnormal beat intervals (e.g. ectopic beats) are not typically marked as noise, since these can be reliably corrected by the automatic beat correction (which is by default performed after noise detection). All segments marked as noise will be excluded from HRV analysis. Please note that you can also manually edit the noise segment markings if necessary, see Fig. 3.

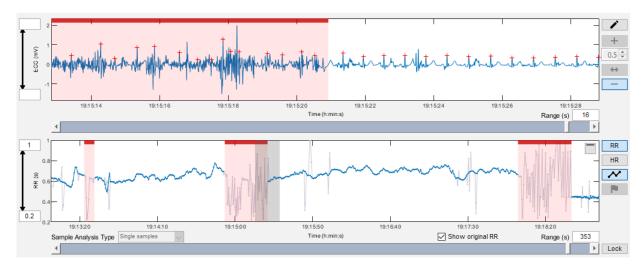


Figure 3: Illustration of noise segment markings in Kubios HRV Premium software. Noise segments can be manually edited from ECG or RR data axes with mouse (right-click to remove an existing noise marking, CTRL + left mouse button (Win) or Command + left mouse button (macOS) to mark/unmark noise).

### 3.3 Artefact correction algorithms

Artefacts in the IBI time series can cause significant distortion to HRV analysis results, and thus, all artefacts should be either corrected or excluded from analysis as recommended also in [30]. Typical artefacts include missing, extra or misaligned beat detections as well as ectopic beats such as premature ventricular contractions (PVC) or other arrhythmias.

Kubios HRV includes two methods for correcting artefacts and ectopic beats present in the IBI data: 1) Threshold based correction, in which the artefacts and ectopic beats are simply corrected by comparing every RR interval value against a local average interval; 2) Automatic correction (only available in Premium), in which artefacts are detected from a time series consisting of differences between successive RR intervals. **The automatic correction** is more accurate and the method has been validated in [14]. In addition to these IBI data based correction methods, the beat detections can be corrected manually when ECG data is used for analysis (see Section 4.2).

The **threshold based artefact correction algorithm** compares every IBI value against a local average interval. The local average is obtained by median filtering the IBI time series, and thus, the local average is not affected by single outliers in IBI time series. If an IBI differs from the locale average more than a specified threshold value, the interval is identified as an artefact and is marked for correction. The threshold value can be selected from:

Very low: 0.45 sec (threshold in seconds)

· Low: 0.35 sec

· Medium: 0.25 sec

· Strong: 0.15 sec

Very strong: 0.05 sec

· Custom, for setting a custom threshold in seconds

3.4 IBI time series trend removal

For example, the "Medium" correction level will identify all IBIs that are larger/smaller than 0.25 seconds compared to the local average. The correction is made by replacing the identified artefacts with interpolated values using a cubic spline interpolation. Please note that the thresholds shown above are when 60 bpm hear rate and are adjusted according to mean heart rate (i.e. lower thresholds for higher heart rate). The correction level should be adjusted individually, because inter-individual difference in HRV are quite significant and therefore a fixed threshold does not work optimally for all subjects. The optimal threshold is the lowest correction level which identifies all artefacts but does not identify too many normal RR intervals as artefacts.

In the **automatic artefact correction algorithm**, artefacts are detected from dRR series, which is a time series consisting of differences between successive RR intervals. The dRR series provides a robust way to separate ectopic and misplaced beats from the normal sinus rhythm. To separate ectopic and normal beats, time varying threshold (Th) is used. To ensure adaptation to different HRV levels, the threshold is estimated from the time varying distribution of the dRR series. For each beat, quartile deviation of the 90 surrounding beats is calculated and multiplied by factor 5.2. Beats within this range cover 99.95% of all beats if RR series is normally distributed. However, RR interval series is not often normally distributed, and thus, also some of the normal beats exceed the threshold. Therefore, decision algorithm is needed to detect artefact beats.

Ectopic beats form negative-positive-negative (NPN) or positive-negative-positive (PNP) patterns to the dRR series. Similarly long beats form positive-negative (PN) and short beats negative-positive (NP) patterns to the dRR series. Only dRR segments containing these patterns are classified as artefact beats. Missed or extra beats are detected by comparing current RR value with median of the surrounding 10 RR interval values (medRR). A missed beat is detected if current RR interval (RR(i)) satisfies condition

$$\left| \frac{RR(i)}{2} - medRR(i) \right| < 2Th \tag{1}$$

and an extra beat is detected if two successive RR intervals (RR(i) and RR(i+1)) satisfy condition

$$|RR(i) + RR(i+1) - medRR(i)| < 2Th.$$
(2)

Detected ectopic beats are corrected by replacing corrupted RR times by interpolated RR values. Similarly too long and short beats are corrected by interpolating new values to the RR time series. Missed beats are corrected by adding new R-wave occurrence time and extra beats are simply corrected by removing extra R-wave detection and recalculating RR interval series.

#### 3.4 IBI time series trend removal

Another common feature that can alter the analysis significantly are the slow linear or more complex trends within the analysed time series. Such slow nonstationarities are characteristic for HRV signals and should be considered before the analysis. The origins of nonstationarities in HRV are discussed, e.g., in [4]. Two kinds of methods have been used to get around the nonstationarity problem. In [32], it was suggested that HRV data should be systematically tested for nonstationarities and that only stationary segments should be analysed. Representativeness of these segments in comparison with the whole HRV signal was, however, questioned in [10]. Other methods try to remove the slow nonstationary trends from the HRV signal before analysis. The detrending is usually based on first order [15, 17] or higher order polynomial [25, 17] models. In addition, Kubios HRV software includes an advanced detrending method originally presented in [29]. This method is based on smoothness priors regularisation and is suitable also for more complex trends of the time series. The method works like a time-varying highpass filter, smoothing the data according to the value of the smoothing parameter. The bigger the smoothing parameter is the lower is the cutoff frequency of the filter. We recommend using the smoothness priors method for removing IBI time series nonstationarities. The cutoff frequency should be below the low frequency band (< 0.04 Hz) not to remove any parts of the normal short term HRV.

#### 4 Kubios HRV user interface

In this section, the graphical user interface (GUI) of Kubios HRV analysis software explained. Please note that functionalities available in the GUI depend on the version you are using (Standard or Premium). All the screen captures shown in this documentation are taken from the Kubios HRV Premium. Features available only in Premium are mentioned in the text.

The user interface window of Kubios HRV is shown in Fig. 4. The user interface is divided into three segments: 1) the RR interval series options segment on the top left corner, 2) the data browser segment on the top right corner, and 3) the results view segment on the bottom. These segments are described in Sections 4.1, 4.2 and 4.3, respectively.



Figure 4: The graphical user interface of Kubios HRV analysis software.

#### 4.1 RR interval series options

The RR interval series options shown in Fig. 5 includes three functions: 1) *Noise segments*, 2) *Beat correction*, and 3) *Samples for analysis*. Noise segment options can be used to mark time periods where data is too noisy to be analyzed. You can adjust the automatic noise detection level (the default level can be changed from preferences) and you can also manually edit noise segment markings. Time periods marked as noise are excluded from HRV analysis (see Section 3.2). The beat correction options can be used to correct artifacts from a corrupted RR interval series. The user can select between two methods: 1) Automatic correction (available only in Premium) and 2) Threshold correction. The automatic correction is an accurate algorithm for detecting artifacts (missed, extra and misaligned beat detections) as well as ectopic beats. The threshold correction simply compares every beat interval against a local mean RR, and identifies the beat as artifact if it exceeds the specified threshold. The threshold should be selected individually, because normal variability in RR intervals can be quite different between

4.2 Data browser

individuals, and therefore, a fixed threshold could over or under correct the RR data. For details on these correction algorithms, see Section 3.3.

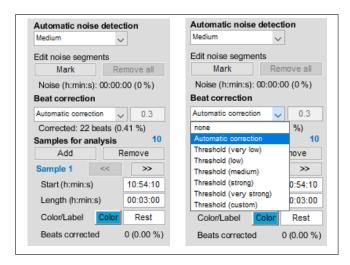


Figure 5: The RR interval series options segment of the user interface.

When using any of the beat correction methods, Kubios HRV displays on the RR data axis the corrected RR interval time series in blue and original uncorrected data in light grey as shown in Fig. 6 A. In this case, the RR interval data includes two clear artifacts: 1) a simulated ectopic beat at 10:13:40 (short interval followed by longer interval) and 2) a simulated missed beat detection at 10:15:30, which are both nicely corrected by using the automatic correction. The number of corrected artifacts in the whole recording is given under the Artifact correction controls and the number of artifacts corrected from the analysis sample is given under Samples for analysis controls. A piecewise cubic spline interpolation method is used in the corrections.

The importance of artifact correction is highlighted in Figs. 6 B-C. Please observe that having only two artifacts within the 5-min analysis segment has a significant effect on the time-domain HRV parameters, especially on SDNN, RMSSD and TINN. Thus, even single artifacts should always be either corrected or excluded from analysis. On the other hand, the number of corrected beats should not be too high (preferably <5%) not to cause significant distortion (suppressed variability) to analysis results. Finally, if ECG is measured, you should first try to directly correct the R-wave detections shown in the ECG data axis as described in Section 4.2.

Using the Samples for analysis options, you can add as many analysis samples that you want (the number of samples is not limited) and you can easily modify the positioning and length of these samples. You can edit the samples using the Add (adding a new sample), Remove (removing the active sample), Start (changing sample onset), Length (changing sample length), Sample Label (an illustrative name for the sample), and Sample Color (e.g. color-code different types of samples) controls. In addition, you can also edit the samples using mouse directly on the RR data axis as described in Section 4.2. Sample Label and Color are optional. By default, Kubios HRV computes analysis results for every selected sample, but you can also choose to merge all samples into one longer sample before analysis using the Sample analysis type control under the RR data axis.

NOTE: In Kubios HRV Premium, you can automatically generate analysis samples by editing desired sample information within a structured CSV file ("Kubios\_Samples.csv" found from the Sample Data folder). In the CSV file you can can define Sample Label, Color, Start time and End time for as many samples as necessary. Analysis samples can be defined differently for every file in the folder or you can also define general sample definitions that will be used for every file in the folder. More information about editing the CSV file is provided on the file header. If the CSV file is placed in the same folder with the data files and sample definitions for a given data file are found, the analysis samples will be automatically imported into Kubios HRV. This feature will make analysis of larger data sets with known sample times much easier.

#### 4.2 Data browser

The data browser segment shown in Fig. 7 displays the measured ECG signal and the extracted RR interval series. If RR interval data is given as input (no ECG data), then only the RR data axis will be displayed in a bigger size. The ECG and RR interval data axes can be controlled by using Sliders (scrolling the data), Range edit boxes (to change data range shown on the axes), and Y-limit edit boxes (to change the y-axis scaling). The range of the ECG axis is

4.2 Data browser

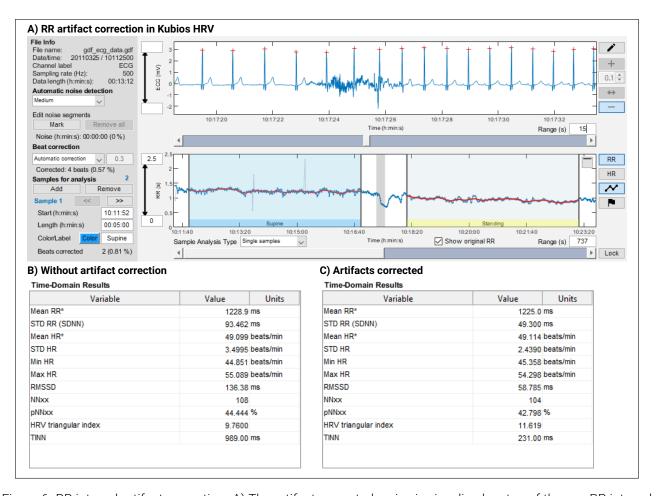


Figure 6: RR interval artifact correction. A) The artifact corrected series is visualized on top of the raw RR interval series, summary of corrected beats within the recording is given on the right side of RR data axis. Time-domain analysis results B) before artifact correction and C) after the artifacts have been corrected.

displayed as a grey patch on the RR axis, which can be moved with left mouse button. ECG and RR axes can also be locked (Lock toggle button) and scrolled together.

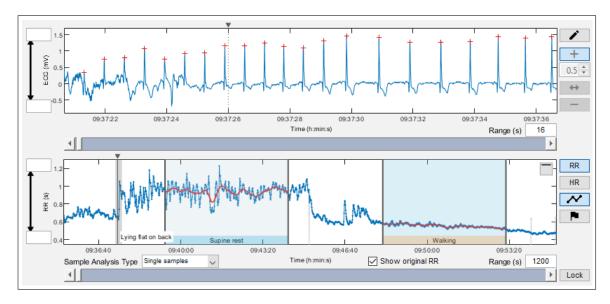


Figure 7: The data browser segment of the user interface.

The main functionalities of the data browser segment are described below.

4.3 Results view 16

**Correcting beat detections** – If ECG (or PPG) data is available, each detected beat is marked in the ECG (or PPG) axis with a "+" mark. Beat detection markers can be easily edited using the tools on the right hand side of the ECG (or PPG) axis. The button actions (from top to bottom) are as follows:

- Turn on/off the beat detection editing mode. When editing beat detections, HRV analysis results are not updated (auto-refresh results turned off) to guarantee smooth editing. When exiting the beat editing mode, any changes made in beat detection become effective and analysis results are updated accordingly.
- Add (shortcut "a") beat detection markers for missed beats.
- Adjust beat detection marker snapping window width (shortcut SHIFT+Scroll) when in Add mode.
- Move/shift (shortcut "s") existing beat detection markers for misaligned beat detections.
- Remove/delete (shortcut "d") beat detection markers for extra beat detections.

**Data markers** – Event markers stored in the recording (if any) are shown both at top of ECG (or PPG) and RR interval data axes as small black triangles. On mouse-over, the text of the marker is displayed. Analysis samples snap to event markers, to ease-up analyzing the data according to available markers. By clicking the "■" button on the right, you can choose which markers are displayed.

**RR data plot options** – From the right side of the RR interval axis, you choose if the data axes shows RR interval or HR values, and if the data is plotted as a dotted line or normal line (""). Also, from the button on top-right corner, you can choose to enlarge the RR data axis.

#### Mouse gestures/shortcuts

- Pan (scroll, SHIFT + scroll) and zoom (Win: CTRL + scroll, maxOS: Command + scroll) data on RR and ECG data views by using mouse scroll wheel.
- Pan both RR and ECG data views by dragging with middle mouse button.
- Add a new analysis sample or noise segment (Win: CTRL + left mouse button + drag, macOS: Command + left mouse button + drag).
- Delete analysis sample or noise marking, or duplicate an analysis sample (right click to open menu).
- Move or resize analysis samples by dragging them with left mouse button from the middle of the sample (to move) or from the left or right edge (to resize).
- Center ECG data view to current cursor position in RR data view (Shift + left mouse click).

#### 4.3 Results view

The results for the selected RR interval sample are displayed in the results view segment (see Fig. 4, which shows results for a maximal cardiopulmonary exercise test). The results are divided into 1) Overview, 2) Time-domain, 3) Frequency-domain, 4) Nonlinear, and 5) Time-varying (available only in Premium) categories. The results of each category are displayed by pressing the corresponding button on the top of the results view segment. The results are by default updated automatically whenever analysis samples or analysis settings are changed. The processing time for computing all the analysis results depends on the length of measurement and number of analysis samples. If the updating of the results takes too much time, you can disable the automatic update by unchecking the "Auto-refresh results" check box on top of the results view segment. When unchecked, you can do all the changes to the analysis samples and settings at once, and then press the "Refresh" button when you want to update the results.

#### 4.3.1 Results overview

The results overview section shown in Fig. 8 displays 1) a comparison between HRV parameters of selected analysis segment and normal resting values and 2) an overview of recording. The normal values for the HRV parameters (Mean RR, Mean HR, RMSSD, SD1 (%) and SD2 (%)) are obtained or derived from the quantitative systematic review by Nunan et al. 2010 [20]. The Stress index is the square root (to make the index normally distributed) of the Baevsky's stress index proposed in [2] and values of Baevsky's stress index between 7 and 12 are considered normal. The six HRV parameters divided into those reflecting paramsympathetic nervous system (PNS) tone (Mean RR, RMSSD and SD1 (%)) and those reflecting sympathetic nervous system (SNS) tone (Mean HR, Stress index

4.3 Results view

and SD2 (%)) are illustrated on top of the normal value distributions. These graphs give a quick view about the level of subject's HRV with respect to normal values. NOTE: the normal values are from rest measurements, thus exercise or stressful situations are expected to produce higher heart rate and lower HRV. PNS and SNS indexes are computed to provide an overall measures of these nervous system activities as compared to normal resting values.

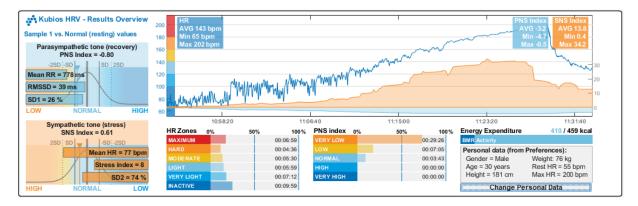


Figure 8: Results overview segment of Kubios HRV.

The overview of the recording includes an illustration of HR, PNS index and SNS index time trends; HR zones graph (time spent in each zone); PNS index zones (time spent in each zone); and energy expenditure (basal metabolic rate and activity related energy expenditure). These information are also provided in the time-varying report sheet (see Section 5.2), where also SNS index zones are illustrated. HR, PNS index and SNS index zones are defined as follows:

	HR zones		PNS zones		SNS zones
	(of HR <sub>max</sub> )				
MAXIMUM:	90-100%	VERY LOW:	< -2	VERY HIGH:	> 2
HARD:	80-90%	LOW:	-21	HIGH:	$1 \dots 2$
MODERATE:	70-80%	NORMAL:	$-1 \dots 1$	NORMAL:	-11
LIGHT:	60-70%	HIGH:	$1 \dots 2$	LOW:	-21
VERY LIGHT:	50-60%	VERY HIGH:	> 2	VERY LOW:	< -2
INACTIVE:	< 50 %				

The total energy expenditure is divided into 1) Basal metabolic rate (BMR) and 2) Activity related energy expenditure (EE). Energy expenditure (EE) is computed using the Keytel's model without a measure of fitness (VO<sub>2</sub>, max) according to [13]. The BMR is estimated using the Mifflin-St Jeor equations [16], which have been found to be the most accurate. The energy expenditure computations are based on heart rate, body weight, height and age, which can be defined in Preferences. In Kubios HRV Premium, these settings can also be changed by clicking the Change Personal Data button (to enable a quick way to update personal details for the current recording).

In Premium version, in addition to energy expenditure, training impulse (TRIMP) is also computed. The TRIMP is computed according to exponential Banister's model [18]. Both EE and TRIMP are computed using beat-to-beat HR values, and thus, their instantaneous values (EE in kcal/min and TRIMP/min) can be reliably derived and these are reported at time-varying results.

#### 4.3.2 Time-domain results view

The time-domain results view shown in Fig. 9, displays the time-domain HRV parameters in a table on the left. The two figure on the right show the RR interval histogram and HR deceleration capacity (DC) and acceleration capacity (AC) graphs. All HRV parameters are calculated from the detrended RR interval data (if detrending is applied), but mean RR, mean HR as well as min and max HR values are exceptions (marked with the \* symbol). In the edit boxes below the RR histogram, you can define fixed lower and upper limits for RR values. These limits are saved in software preferences, so you only need to enter them once. These limits have effect on how the RR histogram is displayed, not only in the results view segment but also in the report figure described in Section 5.2. If you leave the edit boxes empty, the histogram is auto-scaled according to the minimum and maximum values of the data.

4.3 Results view

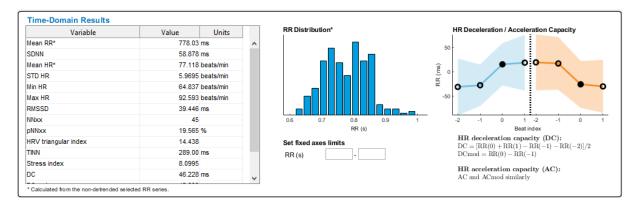


Figure 9: Time-domain HRV analysis results view of Kubios HRV software.

#### 4.3.3 Frequency-domain results view

The frequency-domain results view shown in Fig. 12, displays the results for both FFT and AR spectrum estimation methods. Both methods are applied to the detrended RR series. The spectra of the two methods are presented in the two axes (FFT spectrum on the left and AR spectrum on the right). In Premium version, user can choose in software preferences (see. Section 6) to use Lomb-Scargle periodogram instead of FFT based Welch's periodogram. The frequency axes of the spectra are fixed to range from 0 Hz to the upper limit of HF band plus 0.1 Hz. Thus, for the default frequency band settings the frequency axis range is 0–0.5 Hz. The results for both spectra are displayed in the table on the left. An estimate of respiration rate (RESP) is also computed from the ECG (if available) and RR data. Respiration rate is shown as a vertical line in both spectrum estimates and the numeric value is shown below the spectrum Y-limit options.

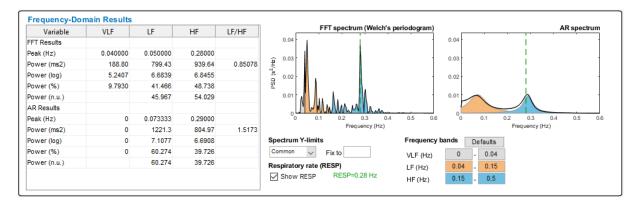


Figure 10: Frequency-domain HRV analysis results view of Kubios HRV software.

The frequency-domain results view includes the following settings. The power axes limits, can be adjusted with the options below the spectrum axes. The power axes can be selected to have either common (same limits for FFT/Lomb and AR spectra) or separate upper Y-limits. If common Y-limit is selected, it can also be entered manually into the edit box beside the selection button. The selected power axis options apply also for the report sheet. Below the spectrum Y-limits options, there is a checkbox, which can be used to show/hide the respiration rate (RESP). In addition, you can find settings for the very low frequency (VLF), low frequency (LF), and high frequency (HF) bands limits. The default values for the bands are VLF: 0-0.04 Hz, LF: 0.04-0.15 Hz, and HF: 0.15-0.4 Hz according to [30]. The default values for the bands can be restored by pressing the Defaults button. Adjustments to the frequency bands here apply only for the current session, if you want to change these settings permanently, you need to do it by editing software preferences (see Section 6).

#### 4.3.4 Nonlinear results view

The nonlinear results view shown in Fig. ??, displays all the calculated nonlinear variables in one table. The Poincaré plot and the DFA results are also presented graphically in the two axes. In the Poincaré plot (left hand axis), the successive RR intervals are plotted as blue dots and the SD1 and SD2 variables obtained from the ellipse fitting

4.4 Menus and toolbar buttons

technique are presented. In the DFA plot (right hand axis), 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  $\alpha_1$  and  $\alpha_2$ , respectively, are indicated.

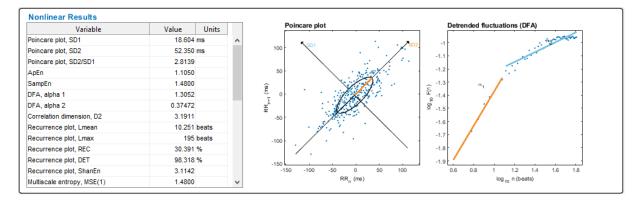


Figure 11: Nonlinear HRV analysis results view of Kubios HRV software.

#### 4.3.5 Time-varying results view

The time-varying results view shown in Fig. ??, displays the time-varying trend of the selected variables. Time-varying analysis is by default applied to the whole measurement, but you can also change Preferences to perform time-varying analysis for every analysis sample (see Section 6). You can either view two selected HRV variables or the time-varying spectrum. Selected variables are plotted in the right hand side axis (plot style can be changed between line plot and filled patch plot from the toggle buttons). You can also choose if you want a background grid and data point markers to be plotted (checkboxes below variable selections). When the time-varying spectrum is selected for view, a color bar indicating the power values is also shown on the right. The color map of the spectrum can be changed with the Color map dropdown button. The adjustable options for the time-varying analysis include the window width and grid interval for the moving window, which is used to calculate the results.

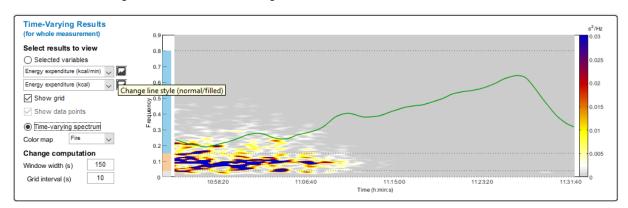


Figure 12: Time-varying HRV analysis results view of Kubios HRV software.

#### 4.4 Menus and toolbar buttons

The user menus and toolbar buttons are located on the upper left hand corner of the user interface. There are all together four user menus and eight toolbar buttons. The toolbar button icons and their actions are explained below

- Open new data file button is for opening a new data file for analysis. If the results of the current analysis have not been saved, user is prompted to do so.
- **Save results** button is for saving the analysis results. The results can be saved in ASCII, PDF, and MATLAB MAT file format (see Section 5 for details).

- Append results to "SPSS friendly" batch file (available only in Premium) button is for adding the current analysis session results into an existing (or creating a new) "SPSS friendly" batch file (see Section 5 for details).
- **Print results** button is for printing the current results without opening report sheet windows.
- **Report preview** button opens the report sheet preview window which include all the analysis results (see Section 5.2 for details).
- **Edit preferences** button opens a preferences window in which you can, e.g., change the default values for analysis options (see Section 6 for details).
- **About Kubios HRV** button opens the about dialog of the software, which includes the version number and contact information. Also the Kubios HRV End User License Agreement can be viewed in the about dialog.
- **Zoom in/out** buttons can be used to zoom in or out on the ECG and RR data axes (please note that in other axes zooming is not enabled).
- Close file button closes the current data file. If the results of the current analysis have not been saved, user is prompted to do so.
- **Show/hide markers** button toggles between showing and hiding event marker time points over the ECG and RR axes.
- **Show/hide marker lines** button toggles between showing and hiding event marker vertical dashed lines over the ECG and RR axes.

All the above actions are also available on the user menus. The **File menu** includes Open, Save Results, Save Results As, Append Results to "SPSS friendly" Batch File, Print Results, Edit Preferences, Close, and Quit commands. The Quit command of the File menu is for exiting from the software. The File menu also displays the last nine opened data files. The **View menu** includes Markers menu, Display ECG/PPG printout preview (an example of ECG printout is shown in Fig. 20), and Report preview command. The Markers menu is for displaying possible stimuli or event markers presented in the experimental procedure and stored in the data file. If no markers are found from the data file the Markers menu will be disabled. The **Tools menu** includes Polar Flow Export option, which provides an easy way to export all your Polar Flow measurements into your computer in FIT file format to be analyzed in Kubios HRV (see Appendix A for details). Finally, the **Help menu** includes links to Kubios HRV User's guide, Kubios home page, Kubios Support pages, Contact us by e-mail, License information, and the About Kubios HRV dialog.

## 5 Saving the results

The analysis results can be saved by selecting Save Results or Save Results As from the File menu or by pressing the save button on the toolbar. This will open a file save dialog in which the saving type can be selected. There are three different types in which the results can be saved: 1) the results can be written in an ASCII text file for further inspection, 2) the report sheets generated from the results can be saved in a PDF-file, and 3) the results can be saved in a MATLAB MAT-file (available only in Premium). In addition to these, Kubios HRV Premium includes the Append to "SPSS friendly" batch file option to save session results into existing batch file (ideal for saving group results).

#### 5.1 ASCII text file

When the ASCII text file is selected for the saving type, the numeric results of the analysis will be written in an CSV formatted text file. The resulting text file includes the following information in the enumerated order.

- 1. Software, user, and data file informations
- 2. Used analysis parameters
- 3. Samples selected for analysis
- 4. Results overview

5.2 PDF report 21

- 5. Time-domain results
- 6. Frequency-domain results
- 7. Nonlinear results
- 8. Time-varying results
- 9. RR interval data and spectrum estimates

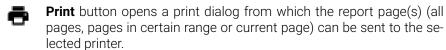
The columns of the file are separated with comma or semicolon (can be adjusted in software preferences) so that the results could easily be imported to, e.g., spreadsheet programs such as the Microsoft Excel<sup>®</sup> for further inspection. A preview of the CSV formatted text file showing standard analysis results is presented in Fig. 21 and showing time-varying results in Fig. 22.

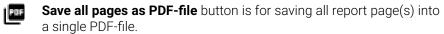
#### 5.2 PDF report

The PDF report includes all the analysis results for the current session. The report pages opens in a preview window where they can be easily printed or saved into a PDF file. Kubios HRV produces one report page for every analysis sample including all the time-domain, frequency-domain, and nonlinear analysis results as shown in Fig. 23. In addition, Kubios HRV Premium produces one page (or several pages if time-varying analysis has been set to apply into analysis samples in Preferences) including time-varying analysis results as shown in Fig. 24. The RR interval data and the sample selected for analysis are presented on top of all report pages and the analysis results below them

When Save Results have been selected, the report page(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 page(s) will not be displayed, but just saved in the selected PDF-file. If you wish to view the report page(s), choose Report preview from the View menu or just press the corresponding toolbar button.

The report preview window includes 11 toolbar buttons and File and Page menus on the upper left hand corners of the windows. The toolbar buttons are explained below





- **Zoom in** button if for zooming in (magnifying) the report page.
- **Zoom out** button is for zooming out the report page.
- Reset to original size button can be used to restore the original zoom level. This also resets the size of the corresponding report preview window to its original size.
- **Move visible area** button is for moving the visible area of the zoomed report page in the report preview window (just grab the sheet with mouse and drag it to the desired direction).
- **Close** button is for closing the report preview.
- **Go to first page** button is for displaying the first report page in the preview.
- **Go to previous page** button is for displaying the previous report page in the preview.
- **Go to next page** button is for displaying the next report page in the preview.
- **Go to last page** button is for displaying the last report page in the preview.

The File menu includes Save All Pages as PDF, Print and Close commands, which are also given as toolbar buttons described above. The Page menu includes commands for changing the page that is displayed in the preview window (First page, Previous page, Next page, Last page), which are all also given as toolbar buttons.

## 5.3 Matlab MAT-file (available in Premium)

In addition to saving the numeric results into an ASCII text file or saving the report sheet(s) in a PDF-file, the analysis results can also be saved in a MATLAB MAT-file (compatible with MATLAB® R2006b or later). The MAT-file include all the analysis results and analysis parameters, exactly as they where when you saved the results. In addition, these files include the raw data (ECG or RR data). This saving option has two purposes:

- 1. The main purpose of the MAT-file is that by opening the MAT-file in Kubios HRV, you can return to the previously performed analyses session as it was (all settings and analysis samples are presented as they were) when the analysis was originally performed. Thus, the MAT file makes it easy for you to change something in the analysis (e.g. add a new analysis sample or change some settings) and re-analyse the data. Thus, we recommend that you save the analysis results always as a MAT-file, just in case if something needs to be done differently.
- 2. In addition, the MAT files are useful for anyone working with MATLAB (further analysis or processing can be performed easily by loading the MAT-files into MATLAB).

The MAT files include a single structured array variable named Res. The Res variable includes the numeric results as well as the RR interval data and all the analysis options. The Res structure includes four fields which are shortly described as follows

f\_name: File name of the analysed data filef\_path: Full path for the analysed data file

CNT: Basic information of the data file (the field name refers to Neuroscan CNT-file

for historical reasons)

HRV: Used analysis options, RR interval data, and all analysis results.

The HRV field is the most essential one of these fields. The HRV field includes six fields the contents of which are shortly described as follows

Param: The analysis options used in the calculation of the results

Data: The RR interval data

Summary: Results overview including PNS and SNS indexes

Statistics: Time-domain analysis results
Frequency: Frequency-domain analysis results
NonLinear: Nonlinear analysis results
TimeVar: Time-varying analysis results

The variable names of the different fields are more or less self-descriptive and are not documented here.

## 5.4 "SPSS friendly" batch file (available in Premium)

In Kubios HRV Premium, you can also save the analysis results into a "SPSS friendly" batch file. This saving options is ideal for saving group results, for example if you need to analyse HRV data of several subjects and want to be able to have the group results easily available for statistical testing e.g. in MS Excel or SPSS. Alternatively, you can use the batch file saving option for saving HRV results of repeated personal recordings, e.g. to monitor training effect or daily stress levels. Kubios uses Comma Separated Values (CSV) file format for the batch file, which can be easily imported into many spreadsheet and statistical software packages (MS Excel, SPSS).

The Append to "SPSS friendly" batch file functions as follows:

- 1. When saving the analysis results of the first subject (i.e. when you want to initialise a new CSV file), select the destination and file name for the new CSV file from the file dialog. In this case, Kubios HRV will initialise the CSV file by writing the column labels and add the analysis results into the first row below the column labels.
- 2. When saving the analysis results of other subjects, simply select the previously saved CSV file. In this case, Kubios HRV will add the results of the current analysis session into the last row of the file.

The structure of the "SPSS friendly" batch file is presented in Fig. 25. Every row of the batch file consists of the file name string and used analysis parameters values ([1x18] array); followed by the following information for every



analysis sample: sample info consisting of sample onset/offset time and artifact correction statistics ([1x2] array), and HRV analysis results ([1x82] array). For more details on the different HRV analysis variables please see Table 3.

## 6 Kubios HRV preferences

Kubios HRV includes several settings related to how the ECG or RR interval data is processed and analysed. The default values for these settings are designed to be suitable for short-term (normal human) HRV recordings, but may sometimes need to be redefined. Some of these settings can be adjusted in the user interface to apply for the current analysis session, but in order to make permanent changes into these settings you need to edit them at software preferences. Preferences can be edited by selecting Edit Preferences from the File menu or by pressing the corresponding toolbar button. This will open the preferences window in which the preference values can be redefined. The preferences are divided into four categories: 1) User information, 2) Input data & pre-processing, 3) Analysis options (divided into time/frequency-domain, nonlinear and time-varying subcategories), and 4) Report settings.

In the **User information** settings shown in Fig. 13 you can set up your personal contact information (Name, Department, and Organization). This information will only be included in the bottom left corner of the report sheet and in the beginning of the ASCII text file including the analysis results. That is, the user information is meant just for indicating the person/organisation that has carried out the analysis. The values given for Gender, Date of birth, Height, Weight, rest HR and Max HR are used in HR zones, training impulse (TRIMP) and energy expenditure computations.

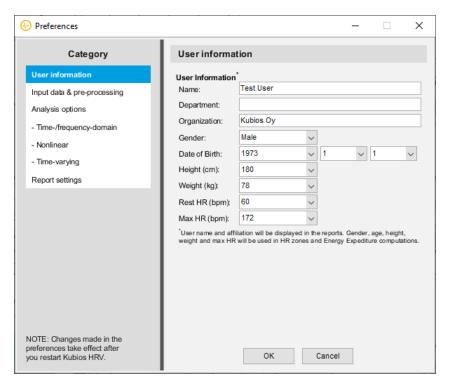


Figure 13: Set up preferences window of the software – User information settings.

From the **Input data & pre-processing** settings shown in Fig. 14, you can change the default input data type into any of the file formats mentioned in Section 2. The selected input data type is used as the default data type every time a new data file is opened, so you can save one additional click by defining the data type you usually work on as the default. The automatic signal quality detection level and default artifact correction method and acceptance threshold can be modified here (see Sections 3.2 and 3.3). By default the acceptance threshold is set to 5%, meaning that analysis samples having more than 5% trigger a warning of low quality data. Under Signal type you can specify if you are using ECG or PPG data for HRV analysis (this has effect on the algorithm applied in beat detection) and if you want to apply an Notch filter to remove power-line noise from the ECG signal (we recommend using the Notch filter only when power-line noise distorts QRS detection). If you are using RR or IBI data, this selection does not have any effect. When ECG is selected as signal type, you can modify QRS detection settings. You can force Kubios to look for the R-waves either from positive of negative amplitudes, or let Kubios to decide (R-wave polarity=Automatic). Also, you can manually fix the prior guess for the average RR interval (used by the QRS detector as initial value), or let Kubios try to estimate it automatically. By default the QRS detection settings are set to automatic and there is no reason to change them unless you are experiencing problems in R-wave detection. If PPG is selected as signal type, you can modify the pulse acceptance threshold (not visible

in Fig. 14 because ECG selected as signal type), which adjusts the sensitivity of the pulse detector algorithm. In addition, the interpolation rate (by default a 4 Hz cubic spline interpolation is applied to form equidistantly sampled time series from the IBI data) and detrending method (by default smoothness priors method is used to remove very low frequency trend components) can be adjusted here. The default detrending settings will remove most of the very low frequency components (frequencies below 0.04 Hz) from the RR interval series prior to analysis. By doing so the HRV analysis results are not affected by the slow changes in HR level and are more sensitive to the short-term HRV regulated by the autonomic nervous system. We recommend removing the trend using the Smoothness priors method which was proposed in [29]. The default settings of the method are optimised for human HRV, but if needed you can change the smoothness of the removed trend by editing the Lambda value. The estimated cut-off frequency for the given Lambda value is presented next to the Lambda value edit box. In the user interface, the trend removed from the RR interval data is shown as a red line over the analysed RR data sample.

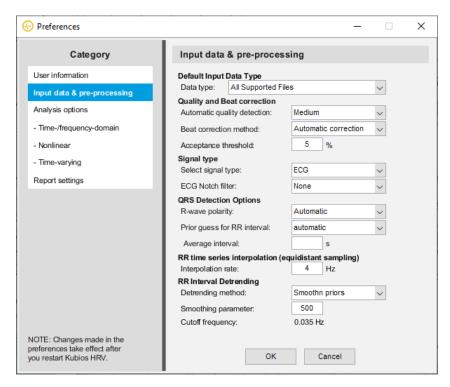


Figure 14: Set up preferences window of the software – Input data & pre-processing settings.

The **Analysis options** settings include some general analysis options, and detailed settings of different analysis methods under three sections: 1) Time/frequency-domain, 2) Nonlinear and 3) Time-varying. The general analysis settings shown in Fig. 15 includes selection of analysis to be performed: 1) Standard analysis (all time-domain, frequency-domain and nonlinear analysis for selected stationary samples), and 2) Time-varying analysis. Only selected analysis will be performed and thus un-checking unnecessary analysis type will speed up the computations. Using the settings for RR interval samples, you can define how many analysis samples are generated by default and what is the length of these samples. Also, the minimum analysis sample length can be defined between 10, 30 and 60 seconds. In case of several samples, you can choose the analysis type between Single samples (in this case, Kubios will perform analysis for every sample separately) and Merge samples (the samples are merged into one longer sample for which analysis is then performed). Finally, the Update mode can be changed between Automatic (analysis results are refreshed automatically) and Manual (you need to refresh results manually).

The time and frequency-domain analysis settings are shown in Fig. 16. For time-domain analysis methods, you can adjust the window width of the moving average filter (default 5 beats), which is used to extract minimum and maximum HR values. Also, you can adjust the threshold used in the computation of NNxx and pNNxx parameters (default 50 ms  $\rightarrow$  NN50 and pNN50). Under HRV frequency bands, the very low frequency (VLF), low frequency (LF), and high frequency (HF) bands of HRV frequency-domain analysis can be adjusted. The default values for these frequency bands are VLF: 0–0.04 Hz, LF: 0.04–0.15 Hz, and HF: 0.15–0.4 Hz according to [30]. The rest of the settings relate to spectrum estimation methods. The points in frequency-domain is given as points/Hz and corresponds by default to the window width of the FFT spectrum. If spectrum interpolation is desired the points in frequency-domain can be increased. The spectrum for the selected RR interval sample is calculated both with Welch's periodogram method (FFT spectrum) and with an autoregressive modeling based method (AR

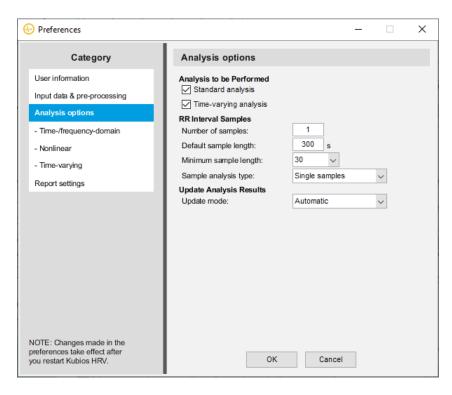


Figure 15: Set up preferences window of the software – Analysis options settings.

spectrum). In the Welch's periodogram method, the used window width and window overlap can be adjusted by editing the corresponding value. The default value for window width is 300 seconds and the default overlap is 50 % (corresponding to 150 seconds), which produce three overlapping windows for a 10-min (600 sec) analysis sample. As an alternative to FFT spectrum, you can select to use the Lomb-Scargle periodogram, which does not assume equidistant sampling and has been recommended for HRV spectral analysis in some studies. The default smoothing window for this spectrum estimate is 0.02 Hz. For the AR spectrum, there are two options that can be selected. First, the order of the used AR model can be selected. The default value for the model order is 16. The second option is whether or not to use spectral factorization in the AR spectrum estimation. In the factorization the AR spectrum is divided into separate components and the power estimates of each component are used for the band powers. Spectral factorisation has been shown to provide some advantage especially when e.g. the HF component is partially overlapping with the LF band [27].

The nonlinear analysis settings are shown in Fig. 17. As the first option, you can choose if the nonlinear parameters are also computed from the detrended RR interval data or not (by default computations are made from detrended data). If you uncheck this option, then all nonlinear parameters are always computed from non-detrended RR interval data. The embedding dimension m (default 2 beats) and the tolerance value r (default 0.2 times SD) used in for the computation of Approximate entropy (ApEn) and Sample entropy (SampEn) can be modified. Note that the tolerance value is adjusted in relation to the standard deviation of the RR interval data. Next, limits of the short-term (N1) and long-term fluctuations used in the Detrended fluctuation analysis (DFA) can be modified (defaults 4-12 and 13-64 beats, respectively). Finally, the embedding dimension (default 10 beats) used both in the computation of the Correlation dimension ( $D_2$ ) and in the Recurrence plot analysis (RPA), and the threshold level (default  $\sqrt{10}$ ) used in RPA, can be modified.

The time-varying analysis settings are shown in Fig. 18. You can choose to apply time-varying analysis (analysis mode) on the whole measurement (default) or for analysis samples. You can adjust the width (default 300 sec) and grid interval (default 60 sec) of the moving window used for time-varying analysis. Note than the analysis window for energy expenditure (EE) and TRIMP can be defined separately, because you may want these estimates to be more instantaneous in nature. The grid interval defines how much the window is moved at every step, and analysis results are available at these intervals. For example, if you want to performe time-varying analysis at 10-min non-overlapping segments for the whole duration of recording, you select "Whole recording" as the analysis mode and define the window width and grid interval both to 600 seconds. Effective data threshold setting adjusts how noise segments affect time-varying analysis results, by default results are not computed unless the effective RR data length (length of data excluding noise) is at least 50% of the analysis window width. For the time-varying spectrum estimation there are two options: 1) the well known spectrogrm (default) and 2) a Kalman smoother

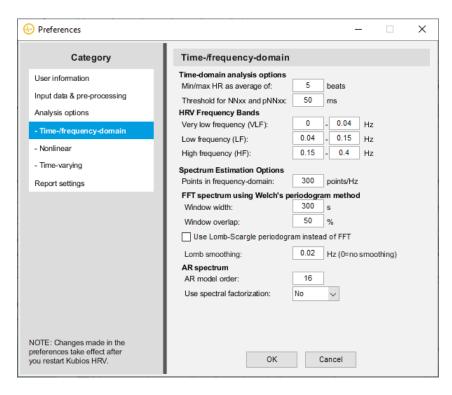


Figure 16: Set up preferences window of the software - Analysis options: time-/frequency-domain methods.

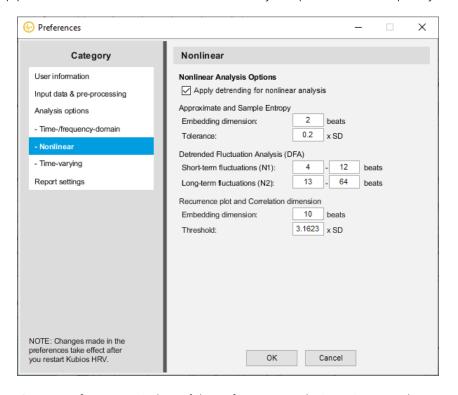


Figure 17: Set up preferences window of the software – Analysis options: nonlinear methods.

spectrum estimate proposed in [27].

The **Report settings** shown in Fig. 19 include the following options. The contents of the results to be exported can be selected by checking the Standard and/or Time-varying results options. If either one of these is unchecked, only the selected results will be exported (in PDF reports or the other export file formats). Concerning the ASCII text file as well as the "SPSS friendly" CSV batch file, the field delimiter and decimal point used when saving the results can be selected (the default values being comma "," for field delimiter and dot "." for decimal separator). The paper size of the report page(s) can be changed between A4 (210×297 mm) and Letter (8.5×11 inch) size.

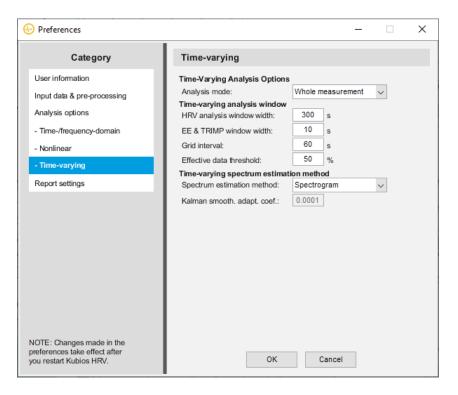


Figure 18: Set up preferences window of the software – Analysis options: time-varying methods.

Only one spectrum figure is shown in the report page, but you can here choose if you want to show the FFT/Lomb or the AR spectrum estimate. For the time-varying report page you can choose four optional HRV parameters to be displayed under time-varying analysis results.

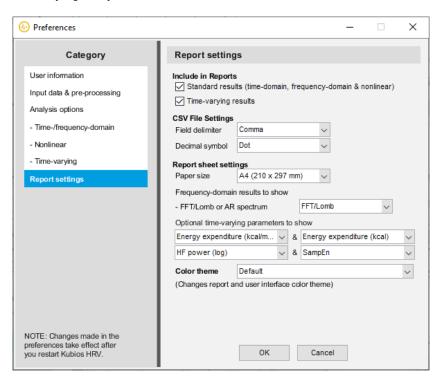


Figure 19: Set up preferences window of the software: Report settings category.

All modifications for the preferences are saved by pressing the OK button. Please note that the changes made into preferences take effect only after restarting Kubios HRV (either closing the analysis session or closing the program). In addition to above described analysis options, there are various other editable options which have mainly influence on the usability of the software. Such options are e.g. the Range and Y-limit values of the data

axis 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.

All the preferences and preserved options used by Kubios HRV are saved in user specific folders<sup>2</sup>.

#### Windows 7,8 or 10:

HRV Standard: C:\Users\<username>\AppData\Roaming\Kubios\KubiosHRVStandard

HRV Premium: C:\Users\<username>\AppData\Roaming\Kubios\KubiosHRVPremium

#### Mac OSX:

HRV Standard: ~/Library/Preferences/Kubios/KubiosHRVStandard

HRV Premium: ~/Library/Preferences/Kubios/KubiosHRVPremium

#### Linux:

HRV Standard: ~/.kubios/KubiosHRVStandard

HRV Premium: ~/.kubios/KubiosHRVPremium

where <username> is the name of your user profile. The folder will include a file named KubiosHRVprefs.mat, which includes all the preferences for the analysis options, user information and user interface usability. The file is created when Kubios HRV is started for the first time and it will be updated whenever the preference values are edited/updated. The original settings of the preferences can be restored by deleting this preference file. Also, if the preference file gets somehow corrupted, the preference file will be rewritten using the default values.

<sup>&</sup>lt;sup>2</sup>Note that the AppData folder in Windows is hidden by default and are not visible in the File Explorer if the "Show hidden files and folders" is not selected from the "Folder Options" section of the File Explorer.

# 7 Kubios HRV analysis parameters

Kubios HRV is provides a detailed analyses of HRV with over 40 analysis parameters. Summary of all HRV parameters calculated by Kubios HRV is given in 3.

Table 3: HRV parameters calculated by Kubios HRV software (\* only preview is available in Standard version; \*\* parameters are available only in Premium version).

Parameter	Units	Description	References
Overview			
Stress index	-	Square root of Baevsky's stress index	[2]
PNS index	-	Parasympathetic nervous system activity compared to normal resting values	
SNS index	-	Sympathetic nervous system activity compared to normal resting values	
HR zones*	[%]	Time spent in Maximum, Hard, Moderate, Light, Very light, and Inactive HR zones	
PNS zones*	[%]	Time spent in Very high, High, Normal, Low and Very Low PNS index levels	
SNS zones**	[%]	Time spent in Very high, High, Normal, Low and Very low SNS index levels	
Energy exp.*	[kcal/min]	Activity related energy expenditure (EE) estimated using Keytel's model	[13]
TRIMP*	[TRIMP/min]	Training impulse (TRIMP) according to Banister's exponetial model	[18]
Time-Domain	[	Training impaids (Training to Barriotor & Experience Training	[.0
RR	[ms]	The mean of RR intervals	
STD RR (SDNN)	[ms]	Standard deviation of RR intervals	
HR	[1/min]	The mean heart rate	
STD HR	[1/min]	Standard deviation of instantaneous heart rate values	A7 F)
Min & Max HR	[1/min]	Minimum and maximum HR computed using <i>N</i> beat moving average (default value:	N = 5
RMSSD	[ms]	Square root of the mean squared differences between successive RR intervals	50)
NNxx	[beats]	Number of successive RR interval pairs that differ more than xx ms (default value: xx	= 50)
pNNxx	[%]	NNxx divided by the total number of RR intervals	Fo o
HRV triangular in-	-	The integral of the RR interval histogram divided by the height of the histogram	[30]
dex	, ,		
TINN	[ms]	Baseline width of the RR interval histogram	[30]
DC, AC**	[ms]	HR deceleration capacity (DC) and acceleration capacity (AC)	[3, 21]
SDANN	[ms]	Standard deviation of the averages of RR intervals in 5-min segments	[30]
SDNNI	[ms]	Mean of the standard deviations of RR intervals in 5-min segments	[30]
Frequency-Domain			
Spectrum		Welch's (or Lomb-Scargle*) periodogram and AR spectrum estimates	
Peak frequency	[Hz]	VLF, LF, and HF band peak frequencies	
Absolute power	[ms <sup>2</sup> ]	Absolute powers of VLF, LF, and HF bands	
Absolute power	[pol]	Natural logarithm transformed values of absolute powers of VLF, LF, and HF bands	
Relative power	[%]	Relative powers of VLF, LF, and HF bands	
riolativo poriol	[.0]	$VLF [\%] = VLF [ms^2]/total power [ms^2] \times 100\%$	
		LF [%] = LF [ms <sup>2</sup> ]/total power [ms <sup>2</sup> ] $\times$ 100%	
		HF [%] = HF [ms <sup>2</sup> ]/total power [ms <sup>2</sup> ] $\times$ 100%	
Normalized power	[n.u.]	Powers of LF and HF bands in normalised units	
Normalizeu power	[II.U.]		
		LF [n.u.] = LF [ms <sup>2</sup> ]/(total power [ms <sup>2</sup> ] – VLF [ms <sup>2</sup> ])	
. ==		HF [n.u.] = HF [ms2]/(total power [ms2] - VLF [ms2])	
LF/HF	-	Ratio between LF and HF band powers	
RESP**	[Hz]	Respiration rate (derived from ECG and RR data)	
Nonlinear			
SD1	[ms]	In Poincaré plot, the standard deviation perpendicular to the line-of-identity	[5, 6]
SD2	[ms]	In Poincaré plot, the standard deviation along the line-of-identity	
SD2/SD1	-	Ratio between SD2 and SD1	
ApEn	-	Approximate entropy	[26, 9
SampEn	-	Sample entropy	[26]
DFA, $\alpha_1$	-	In detrended fluctuation analysis, short term fluctuation slope	[23, 24
DFA, $\alpha_2$	-	In detrended fluctuation analysis, long term fluctuation slope	[,
$D_2^{**}$	-	Correlation dimension	[11, 12]
RPA**:		Recurrence plot analysis:	[31, 8, 33
	[hooto]	Mean line length	[0.,0,00
Lmean	[beats]	Maximum line length	
Lmax	[beats]	9	
REC	[%]	Recurrence rate	
DET	[%]	Determinism Shappan entrany	
ShanEn	-	Shannon entropy  Multipeals entropy for each factor values = 1.2 and 20	r→.
MSE**	-	Multiscale entropy for scale factor values $ au=1,2,\ldots,20$	[7]
Time-Varying**		DNO is deed ONO is deed EF TDIAND and DEOD	
		PNS index, SNS index, EE, TRIMP and RESP	
		, HR, STD HR, Min HR, Max HR, RMSSD, NNxx, pNNxx, HRV tri ind., TINN	
		e-varying spectrum (Spectrogram); VLF, LF and HF peak frequencies;	
VI F I F and HF nov	wers in $ms^2$ . loa	and %; LF and HF powers in n.u.; LF/HF ratio	
VLI, LI dila ili pot		2/SD1, ApEn, SampEn, DFA $lpha_1$ and $lpha_2$	

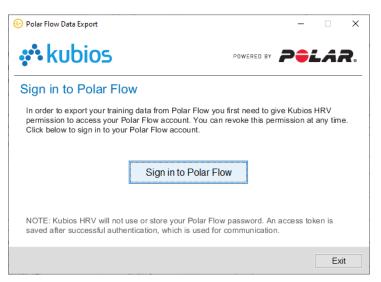
## A Appendix: Polar Flow export (available in Premium)

The Polar Flow export option provides an easy way to export your Polar Flow measurements into your computer hard drive in FIT file format, which can then be analyzed in Kubios HRV software. Previously, Polar users were able to export RR data from Polar Flow only when using the RR recording test available in Polar V800. However, by using the new Polar Flow export functionality, you will have access to the RR data required for HRV analysis from all Polar Flow compatible Polar devices when using them with H6, H7 or H10 heart rate sensor (**NOTE: RR data is not available when measuring with the Polar Beat App**). Furthermore, you can access the RR data also from your exercise mode measurements (which has not been possible before even with the V800 monitor). In order to start exporting your Polar Flow measurements, click Polar Flow Export in Tools menu and follow given instructions. The steps needed to link your Polar Flow account with Kubios HRV are shortly described below. Please note that only those training sessions that are added to Polar Flow after linking with Kubios HRV are accessible through Polar Flow API.

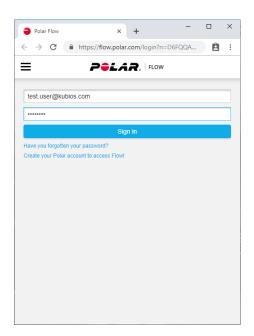
#### Please remember that:

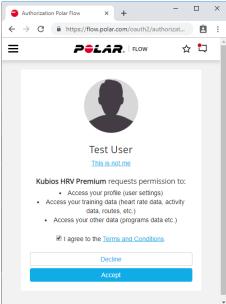
- 1. ONLY MEASUREMENTS MADE AFTER LINKING YOUR POLAR FLOW ACCOUNT ARE ACCESSIBLE
- 2. RR data is available only when a chest strap (H6, H7 or H10) has been used
- 3. Each measurement can be exported only once (take backup copies of exported data files).

When you click the Polar Flow Export option for the first time, a window instructing you to sign in to your Polar Flow account pops up



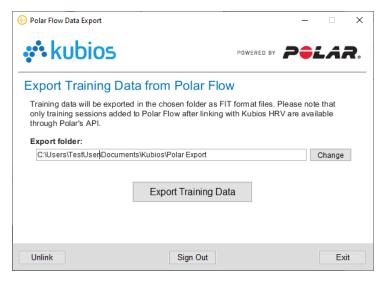
Click "Sign in to Polar Flow" to continue.



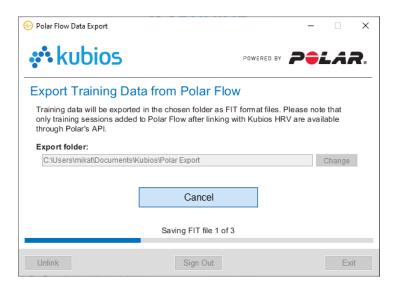


Next, you need to Sign In to your Polar Flow account and give Kubios HRV permission to access your Polar Flow account data. To continue, confirm that you agree to the Terms and Conditions and click Accept. You should then receive a notice that your Polar Flow account is successfully linked with Kubios HRV.

Once you have linked your Polar Flow account with Kubios HRV, you can export your training data by clicking the Polar Flow Export from Tools menu.



You can define the export folder, where your training data from Polar Flow will be exported. Every training measurement that you have uploaded into Polar Flow after linking your account with Kubios HRV or after the previous successful export, will be exported into the selected folder in FIT file format when clicking Export Training Data button.



If you do not have new measurements uploaded to Polar Flow, you will receive a notification "No new training data available in Polar Flow". Please note, that not all the FIT files exported from Polar Flow necessarily include RR data and can not thus be opened in Kubios HRV. RR data is available only when you have used a chest strap (Polar H6, H7 or H10) to record heart rate. RR data is not available when you have measured heart rate from your wrist (wrist-based HR) or by using the optical Polar OH1 armband.

You can Unlink Kubios HRV from your Polar Flow account whenever you want. However, you should do this only if you do not anymore need to access your training data in Kubios HRV. If you want to temporarily disable the access to your Polar Flow account from Kubios HRV, you can Sign Out from your account instead. In this case, you are required to sign in again the next time you want to export data from Polar Flow, but all your latest training data are still accessible through the Polar Flow link.

## **B** Appendix: Kubios HRV figures

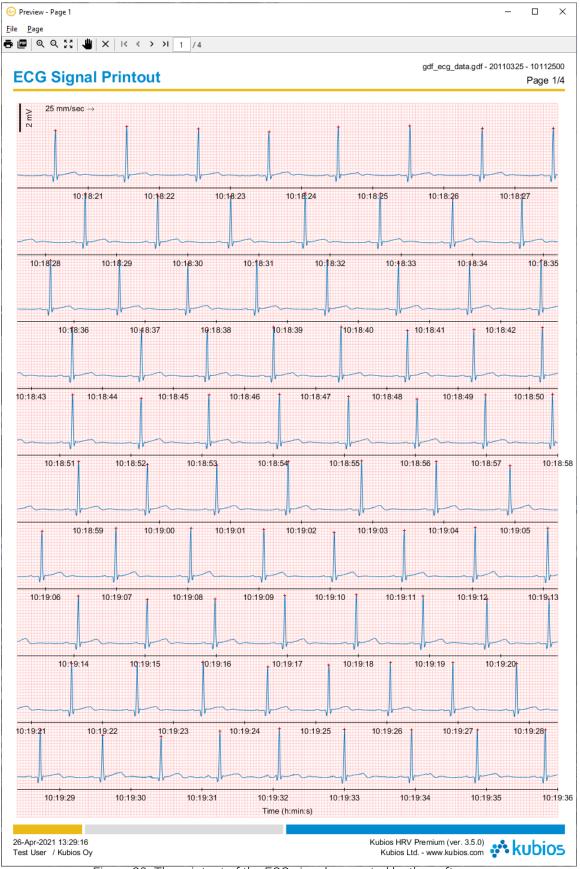


Figure 20: The printout of the ECG signal generated by the software.

4	A	В	С	D	E	F	G	Н	I	J
	Interval Samples Selected for Analys	sis								
1		Rest(1)	Warm-up(2)	40W(3)	80W(4)	120W(5)	160W(6)	200W(7)	240W(8)	Peak(9)
2 Sa	ample limits (hh:mm:ss):	10:54:10-10:57:	10:59:56-11:02	11:04:09-11:07:	11:07:10-11:10:	11:10:11-11:13:	11:13:12-11:16:	11:16:13-11:19:	11:19:14-11:22:	11:25:14-11:28
	ample Analysis Type: Single samples	;								
4 Be	eat correction: Automatic correction									
5 Be	eats total:	231	290	310	338	379	429	477	522	572
5 Be	eats corrected:	0	2	4	2	0	0	2	0	2
7 Be	eats corrected (%):	0	0.69	1.29	0.592	0	0	0.419	0	0.35
B Eff	fective data length (s):	180	180	180	180	180	180	180	180	180
9 Eff	fective data length (%):	100	100	100	100	100	100	100	100	100
)	5 , ,									
1										
_	SULTS FOR SINGLE SAMPLES									
_	ANDARD RESULTS									
1	NIVERIED RESOLTS	Rest(1)		Warm-up(2)		40W(3)		80W(4)		120W(5)
	sults Overview	NESU(1)		warm-up(2)		4000(3)		80W(4)		12000(3)
	NS index:	-0.8		-1.8674		-2.3531		-2.9086		-3.51
-	NS Index:	0.6073		2.3119		3.384		-2.9086 5.9657		8.7605
-										
_	ress index:	8.0995		10.5646		14.1578		25.2804		33.7775
)										
_	ne-Domain Results									
	atistical parameters									
_	lean RR (ms):	778.0303		620.5406		579.744		531.5622		474.682
	ONN (ms):	58.8781		51.521		38.5571		20.952		9.9782
M	lean HR (beats/min):	77.1178		96.6899		103.494		112.8748		126.4004
SD	O HR (beats/min):	5.9695		8.0154		6.5231		4.3521		2.5703
М	lin HR (beats/min):	64.8368		73.0105		78.7712		99.2392		106.5719
	lax HR (beats/min):	92,5926		110.9262		119,7366		123,3046		139.860
_	MSSD (ms):	39.4455		28.6342		19.3617		10.1272		4.362
	Nxx (beats):	45		21		10.5017		10.1272		4.502
_	NNxx (%):	19.5652		7.2664		3,2362		0.2967		(
						3.2362 NaN				
_	DANN (ms):	NaN		NaN				NaN		NaN
	ONN index (ms):	NaN		NaN		NaN		NaN		NaN
	ometric parameters									
	R tri index:	14.4375		9.666667		8.857143		5.451613		2.746377
	NN (ms):	289		234		192		94		65
	C (ms):	46.2283		37.9963		28.8461		12.6265		3.8611
7 DC	Cmod (ms):	42.8331		33.1038		21.1257		9.9574		3.5325
3 AC	C (ms):	-46.0219		-31.7911		-24.4101		-10.2447		-4.7206
) AC	Cmod (ms):	-42.7762		-27.9293		-18.7914		-8.875		-4.0763
0										
l Fre	quency-Domain Results	FFT spectrum	AR spectrum	FFT spectrum	AR spectrum	FFT spectrum	AR spectrum	FFT spectrum	AR spectrum	FFT spectrum
_	ak frequencies									
_	LF (Hz):	0.033333	0	0.026667	0	0.04	0	0.03	0	0.033333
	(Hz):	0.093333	0.1	0.026667	0.093333	0.093333	0.093333	0.093333	0.093333	0.086667
-	(Hz):	0.093333	0.1	0.0000.	0.093333	0.253333	0.276667	0.093333	0.286667	0.196667
_		0.15	0.2	0.173555	0.26	0.233333	0.2/000/	0.16	0.200007	0.130007
	solute powers	255.2522	_	47.00==	907.0000	22.222	240 4202	50.500	0	40.470
_	F (ms^2):	255.2539	0		327.9902	33.3018	249.1382	59.609		13.178:
	(ms^2):	2323.1262	2899.4314	2234.5719	2410.7425	429.3865	1288.3253	451.9943	451.7297	83.054
_	F (ms^2):	946.442	617.2272	646.3698	190.2333	165.0663	66.6487	76.6464	47.8106	18.4172
	.F (log):	5.5423	0		5.793	3.5056	5.518	4.0878	0	2.5786
	(log):	7.7507	7.9723	7.7118	7.7877	6.0624	7.1611	6.1137	6.1131	4.419
HF	F (log):	6.8527	6.4252	6.4714	5.2483	5.1063	4.1994	4.3392	3.8672	2.9133
Re	lative powers									
4 VL	F (%):	7.2416	0	1.6185	11.1982	5.3049	15.5312	10.1333	0	11.4941
_	(%):	65.9076	82.4485	76.3085	82.3069	68.4001	80.3139	76.8371	90.4291	72.4409
_	F (%):	26.8508	17.5515	22.0729	6.4949	26.2946	4.1549	13.0296	9.5709	16.0636
	ormalized powers	20.0300	17.5515	22.0,25	0.4545	20.2340	4.2545	10.0250	5.57.05	10.000
_	(n.u.):	71.053	82,4485	77.5639	92.6861	72 2319	95.0812	85.5012	90.4291	
_		71.053 28.947		22.436		72.2319 27.7676	95.0812 4.9188	14.4988	90.4291	
	F (n.u.):		17.5515 3516.6586	22.436	7.3139 2928.9659	27.7676 627.7575	4.9188 1604.1123	14.4988 588.2501	9.5709 499.5403	114.6514
_	tal power (ms^2):	3524.8221								

Figure 21: Kubios HRV Premium results for exercise test saved as a CSV formatted text file, showing time-domain, frequency-domain and nonlinear HRV analysis results for different exercise protocol time periods (Rest, Warm-up, 40W load, 80W load, and 120W load).

Α	В	С	D	E	F	G	Н	ı	J	K	L	М	
TIN	ME-VARYING RESUL	те											
IIIN	WHOLE DATA	15											
	WHOLE DATA						Overview						
	Time	Poste total	Posts corrected	Beats corrected	Effective data le	Effective data le	PNS index	CNC index	Stress index	EE petivity	EE activity	Intoneity	
	(hh:mm:ss)	(count)	(count)	(%)	(sec)		PNS muex	SNS Index	Stress maex	(kcal/min)		Intensity (TDIAD (asia)	
		184				(%) 100	0.5403	0.3922	0.1022		(kcal)	(TRIMP/min)	
-	10:54:19				150		-0.5403		8.1832	2.6598	0.4433	0.1495	
	10:54:29	186			150	100	-0.5999	0.4109	8.1212	1.6596	0.7199	0.0998	
	10:54:39	186			150	100	-0.6008	0.431	8.1237	2.3559	1.1125	0.1336	
	10:54:49	185	0		150	100	-0.5823	0.4789	8.5857	2.2999	1.4959	0.1307	
	10:54:59	186			150	100	-0.5423	0.4937	8.5679		1.7617	0.0969	
	10:55:09	189	0		150	100	-0.6248	0.5473	8.516		2.0444	0.1015	
	10:55:19	190			150	100	-0.6917	0.5525	8.2368		2.3089	0.0965	
	10:55:29	191	0		150	100	-0.7387	0.5846	8.2231	1.7143	2.5946	0.1023	
	10:55:39	191	0		150	100	-0.7428	0.6289	8.5534	2.9237	3.0819	0.1639	
	10:55:49	192	0		150	100	-0.7746	0.6077	8.2225	2.447	3.4897	0.1383	
	10:55:59	193	0		150	100	-0.7569	0.5354	7.6033	1.6997	3.773	0.1017	
	10:56:09	195	0	0	150	100	-0.8039	0.626	7.8372	2.545	4.1971	0.1434	
	10:56:19	196	0	0	150	100	-0.8316	0.7156	8.2385	3.201	4.7306	0.1798	
	10:56:29	198	0	0	150	100	-0.8575	0.7928	8.4936	2.5276	5.1519	0.1425	
	10:56:39	198	0		150	100	-0.9264	0.8074	8.5231	2.4329	5.5574	0.1375	
	10:56:49	197	0		150	100	-0.9399	0.7967	8.4148	2.3327	5.9462	0.1324	
	10:56:59	197	0		150	100	-0.9089	0.6794	7.8465	2.6052	6.3804	0.1466	
	10:57:09	196	0		150	100	-0.9301	0.7546	8.3936	3.1819	6.9107	0.1787	
	10:57:19	197	0	0	150	100	-0.9609	0.7287	8.1172	4.0155	7.5799	0.2303	
	10:57:29	198	0	0	150	100	-1.0327	0.7735	8.231	2.5115	7.9985	0.1416	
	10:57:39	197	0	0	150	100	-0.9645	0.6562	7.5966	2.9803	8.4952	0.1671	
	10:57:49	197	0		150	100	-0.9869	0.7073	7.9198		8.832	0.1168	
	10:57:59	200			150	100	-1.0652	0.7451	7.6388		9.1304	0.1059	
	10:58:09	205	3		150	100	-1.1912	0.945	8.1151	2.0006	9.4638	0.1159	
	10:58:19	210			150	100	-1.2933	1.0922	8.3333	1.8863	9.7782	0.1104	
	10:58:29	213	3		150	100	-1.4049	1.3307	9.3416		10.1621	0.1309	
	10:58:39	214			150	100	-1.4541	1.4205	9.674		10.6461	0.1629	
	10:58:49	217	5		150	100	-1.5491	1.4648	9.3589		11.0391	0.1023	
	10:58:59	217	5		150	100	-1.6086	1.6464	10.0509	2.3376	11.5041	0.1566	
		219	5		150							0.1500	
	10:59:09					100	-1.6396	1.6779	9.8102	5.1427	12.3612		
	10:59:19	225	5		150	100	-1.5874	1.6251	9.0598	6.7288	13.4827	0.4494	
	10:59:29	228			150	100	-1.6648	1.7235	9.0767	6.5404	14.5727	0.4313	
	10:59:39	232			150	100	-1.7038	1.8044	9.0541	6.0142	15.5751	0.3832	
	10:59:49	235	5		150	100	-1.7845	2.0343	9.8741	4.9726	16.4038	0.298	
	10:59:59	237	5		150	100	-1.8109	2.1058	9.9633	5.6719	17.3492	0.3538	
	11:00:09	240			150	100	-1.9415	2.3504	10.8696		18.2729	0.343	
	11:00:19	243			150	100	-1.9862	2.432	10.915		19.0052	0.2559	
	11:00:29	243	2		150	100	-1.8752	2.4382	11.1015	3.9853	19.6694	0.2283	
	11:00:39	241	2		150	100	-1.8242	2.3102	10.6845		20.5042	0.3007	
	11:00:49	239	2		149	99.3333	-1.7791	2.1345	9.9009	4.7694	21.2991	0.2828	
	11:00:59	239	2		150	100	-1.8044	2.1233	9.7891	5.7222	22.2528	0.358	
	11:01:09	240	2	0.8333	150	100	-1.8249	2.2863	10.5469	4.7845	23.0502	0.2839	
	11:01:19	241	0	0	150	100	-1.8363	2.2404	10.2034	5.6351	23.9894	0.3507	
	11:01:29	241	0	0	150	100	-1.813	2.1887	9.9231	5.353	24.8816	0.3277	
	11:01:39	242	0	0	150	100	-1.8428	2.3299	10.6263	5.0264	25.7193	0.3021	
	11:01:49	244			150	100	-1.9775	2.7174	12.7276		26.4725		
	11:01:59	244			150	100	-1.9693	2.7117	12.7303		27.2895		
	11:02:09	244			150	100	-1.9919	2.7087	12.6988				
	11:02:19	243			150	100	-1.9661	2.5106	11.6267		29.4292	0.4399	
	11:02:29	242			150	100	-1.9145	2.4563	11.4806		30.4224		
	11:02:39	242			150	100	-1.8635	2.4303	10.7499		31.3089		
	11:02:49	239			150	100	-1.8515	2.2761	10.7499		32.1979	0.3249	
	11:02:59	239			150	100	-1.8216	2.1129	10.3699				
		237			150	100					33.1759		
	11:03:09						-1.8667	2.2894	10.9436		34.0016		_
_	11:03:19	239	4	0.8368	150	100	-1.8763	2.3629	11.2341	4.9662	34.8293	0.2975	-

Figure 22: Kubios HRV Premium time-varying analysis results saved as a CSV formatted text file. In this case, time-varying results were computed at 120-sec window with 10-sec grid interval. Note: only the first few columns of results are shown.

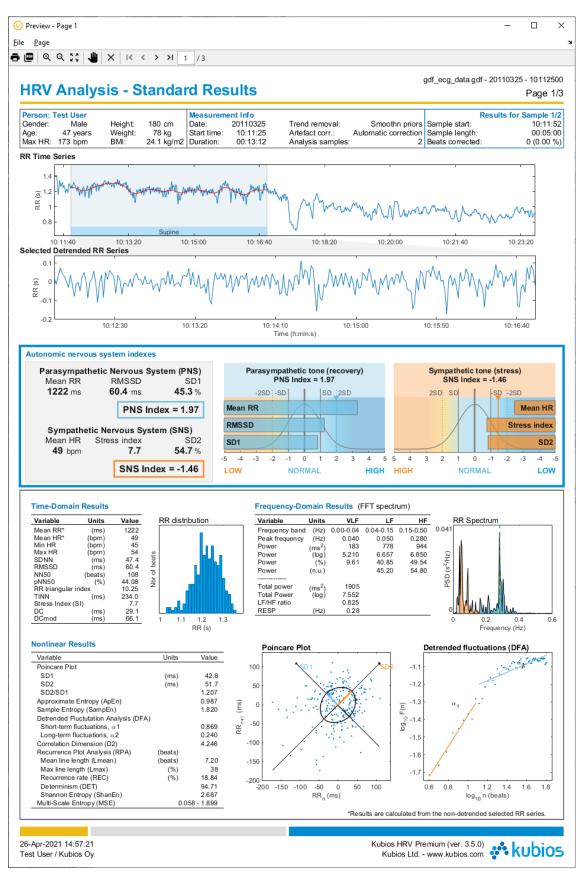


Figure 23: Kubios HRV Premium report page showing HRV analysis results for healthy young male during supine rest (5-min analysis sample).

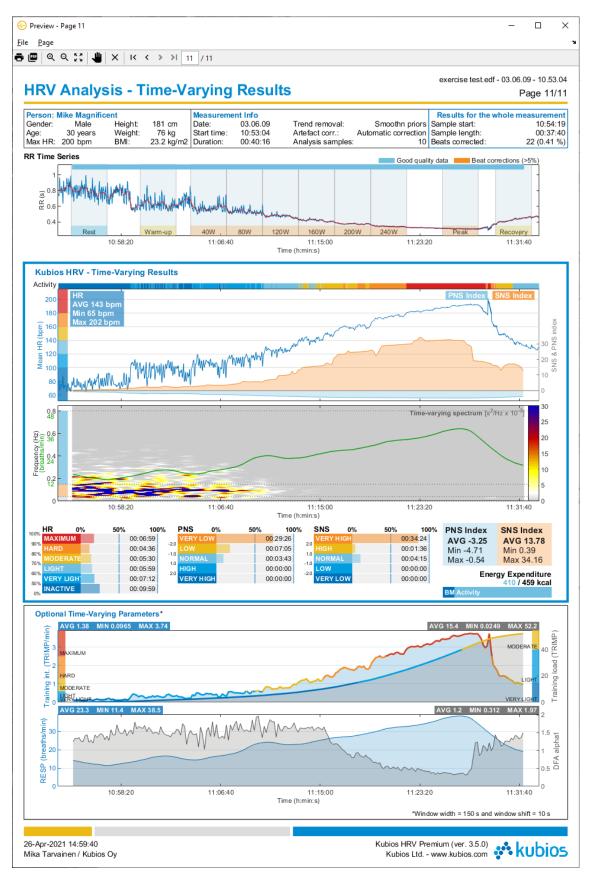


Figure 24: Kubios HRV Premium report page showing time-varying analysis results for maximal cardiopulmonary exercise test performed by healthy young male.

- 1	N	ı	٦

		Sample 1 (S <sub>1</sub> )			Sam	ple $N(S_N)$
Files analysed	Parameters	Info	HRV results		Info	HRV results
FileName	PRM#[PARAMETERS]	S1_[INFO]	S1_[VARIABLES]		S1_[INFO]	S1_[VARIABLES]
subject_1.txt	[1x18] array	[1x2]	[1x82]		[1x2]	[1x79]
subject_2.txt	[1x18] array	[1x2]	[1x82]		[1x2]	[1x79]
:	:	:	:		:	:
:	•	:	:			:
subject_M.txt	[1x18] array	[1x2]	[1x82]		[1x2]	[1x79]

#### **Parameters**

**#Detrending**: Detrending method used #InterpRate: Interpolation rate of RR data #MinMaxHR: Nbr of beats averaged for Min/Max HR #NNxxThreshold: Threshold for NNxx and pNNxx in msec #VLFband: VLF frequency band limits in Hz #LFband: LF frequency band limits in Hz

#HFband: HF frequency band limits in Hz **#FreqPoints**: Nbr of points in spectra (points/Hz) #FFTorLomb: FFT (Welch) or Lomb periodogram used

#### Sample Info

Onset-Offset: Sample onset-offset times (hh:mm:ss)

#### **HRV** variables

PNS index: Parasympathetic nervous system tone index SNS index: Sympathetic nervous system tone index Stress index: Square root of Baevsky's stress index Mean RR (ms): Mean of RR intervals SDNN (ms): Standard deviation of RR intervals

Mean HR (bpm): Mean heart rate SD HR (bpm): Standard deviation of heart rate Min HR (bpm): Minimum HR using N beat MA Max HR (bpm): Maximum HR using N beat MARMSSD (ms): RMS of successive RR interval differences

NNxx (beats): Nbr or successive RRs > xx ms pNNxx (%): Percentage of successive RRs > xx ms

HRV triangular index: RR histogram area/height TINN (ms): RR histogram baseline width DC (ms): HR deceleration capacity

DCmod (ms): Modified HR deceleration capacity

AC (ms): HR acceleration capacity

ACmod (ms): Modified HR acceleration capacity SDANN (ms): SD of 5-min RR interval segment means SDNNI (ms): Mean of 5-min RR interval segment SDs VLFpeak\_FFT\* (Hz): VLF band peak frequency (FFT)

LFpeak\_FFT (Hz): LF band peak frequency (FFT) HFpeak\_FFT (Hz): HF band peak frequency (FFT) VLFpow\_FFT (ms2): Absolute VLF power (FFT) LFpow\_FFT (ms2): Absolute LF power (FFT) HFpow\_FFT (ms2): Absolute HF power (FFT)

VLFpow\_FFT (log): Log VLF power (FFT) LFpow\_FFT (log): Log LF power (FFT) HFpow\_FFT (log): Log HF power (FFT) VLFpow\_FFT (%): Relative VLF power (FFT)

LFpow FFT (%): Relative LF power (FFT) HFpow\_FFT (%): Relative HF power (FFT) LFpow\_FFT (n.u.): Normalised LF power (FFT)

HFpow\_FFT (n.u.): Normalised HF power (FFT) TOTpow\_FFT (ms2): Total spectral power (FFT) LF\_HF\_ratio\_FFT: LF/HF power ratio (FFT)

\* If Lomb-Scargle periodogram is used instead of Welch's periodogram,  $FFT \rightarrow Lomb$ 

#WelchWindow: Window width (overlap) in Welch

#LombWindow: Smoothing window width in Lomb periodogram

#ARspectrum: Order of AR spectrum (factorisation)

**#Entropy**: Embedding dimension (tolerance)

#DFAshortterm: DFA, short-term fluctuations range **#DFAlongterm**: DFA, long-term fluctuations range

#RecurrencePlot: RPA, embedding dimension (threshold)

#NbrSamples: Number of analysed samples

#ArtifactCorrection: RR artifact correction method

Beats corrected (%): Corrected beats within the sample

VLFpeak\_AR (Hz): VLF band peak frequency (AR spectrum) LFpeak\_AR (Hz): LF band peak frequency (AR spectrum) HFpeak\_AR (Hz): HF band peak frequency (AR spectrum) VLFpow\_AR (ms2): Absolute VLF power (AR spectrum) LFpow\_AR (ms2): Absolute LF power (AR spectrum) HFpow\_AR (ms2): Absolute HF power (AR spectrum) VLFpow\_AR (log): Log VLF power (AR spectrum) LFpow\_AR (log): Log LF power (AR spectrum) HFpow\_AR (log): Log HF power (AR spectrum) VLFpow\_AR (%): Relative VLF power (AR spectrum) LFpow\_AR (%): Relative LF power (AR spectrum) HFpow\_AR (%): Relative VLF power (AR spectrum) LFpow\_AR (n.u.): Normalised LF power (AR spectrum) HFpow\_AR (n.u.): Normalised HF power (AR spectrum) TOTpow\_AR (ms2): Total spectral power (AR spectrum) LF\_HF\_ratio\_AR: LF/HF power ratio (AR spectrum)

RESP (Hz): Respiration rate

SD1 (ms): Poincaré plot short term variability SD2 (ms): Poincaré plot long term variability

SD2\_SD1\_ratio: SD2/SD1 ratio ApEn: Approximate entropy SampEn: Sample entropy D2: Correlation dimension

DFA1: DFA, short term fluctuations slope DFA2: DFA, long term fluctuations slope RP\_Lmean (beats): RPA, mean line length RP\_Lmax (beats): RPA, maximum line length

RP\_REC (%): RPA, recurrence rate RP DET (%): RPA, determinism RP\_ShanEn: RPA, Shannon entropy

MSE\_1 ...MSE\_20: Multiscale entropy for scales  $\tau=1,\ldots,20$ 

Figure 25: Structure of the "SPSS friendly" batch file: A) overview of the file structure and B) short description of the fields.

References 40

#### References

[1] F. Badilini, for the ISHNE Standard Output Format Task Force. The ISHNE Holter standard output file format. *A.N.E*, 3(3):263–266, 1998.

- [2] R. M. Baevsky. Methodical recommendations use kardivar system for determination of the stress level and estimation of the body adaptability standards of measurements and physiological interpretation. 2009.
- [3] A. Bauer, J.W. Kantelhardt, P. Barthel, R. Schneider, T. Mäkikallio, K. Ulm, K. Hnatkova, A. Schöming, H. Huikuri, A. Bundle, M. Malik, and G. Schmidt. Deceleration capacity of heart rate as a predictor of mortality after myocardial infarction: cohort study. *Lancet*, 367:1647–81, 2006.
- [4] G.G. Berntson, J.T. Bigger Jr., D.L. Eckberg, P. Grossman, P.G. Kaufmann, M. Malik, H.N. Nagaraja, S.W. Porges, J.P. Saul, P.H. Stone, and M.W. Van Der Molen. Heart rate variability: Origins, methods, and interpretive caveats. *Psychophysiol*, 34:623–648, 1997.
- [5] M. Brennan, M. Palaniswami, and P. Kamen. Do existing measures of Poincaré plot geometry reflect nonlinear features of heart rate variability. *IEEE Trans Biomed Eng*, 48(11):1342–1347, 2001.
- [6] S. Carrasco, M.J. Caitán, R. González, and O. Yánez. Correlation among Poincaré plot indexes and time and frequency domain measures of heart rate variability. *J Med Eng Technol*, 25(6):240–248, November/December 2001.
- [7] M. Costa, A.L. Goldberger, and C.-K. Peng. Multiscale entropy analysis of biological signals. *Physical Rev E*, 71:021906, 2005.
- [8] H. Dabire, D. Mestivier, J. Jarnet, M.E. Safar, and N. Phong Chau. Quantification of sympathetic and parasympathetic tones by nonlinear indexes in normotensive rats. *amj*, 44:H1290–H1297, 1998.
- [9] Y. Fusheng, H. Bo, and T. Qingyu. Approximate entropy and its application in biosignal analysis. In M. Akay, editor, *Nonlinear Biomedical Signal Processing: Dynamic Analysis and Modeling*, volume II, chapter 3, pages 72–91. IEEE Press, New York, 2001.
- [10] P. Grossman. Breathing rhythms of the heart in a world of no steady state: a comment on Weber, Molenaar, and van der Molen. *Psychophysiol*, 29(1):66–72, January 1992.
- [11] S. Guzzetti, M.G. Signorini, C. Cogliati, S. Mezzetti, A. Porta, S. Cerutti, and A. Malliani. Non-linear dynamics and chaotic indices in heart rate variability of normal subjects and heart-transplanted patients. *Cardiovascular Research*, 31:441–446, 1996.
- [12] B. Henry, N. Lovell, and F. Camacho. Nonlinear dynamics time series analysis. In M. Akay, editor, *Nonlinear Biomedical Signal Processing: Dynamic Analysis and Modeling*, volume II, chapter 1, pages 1–39. IEEE Press, New York, 2001.
- [13] L.R. Keytel, J.H. Goedecke, T.D. Noakes, H. Hiilloskorpi, R. Laukkanen, L. Van Der Merwe, and E.V. Lambert. Prediction of energy expenditure from heart rate monitoring during submaximal exercise. *J Sports Sci*, 23(3):289–297, 2005.
- [14] J.A. Lipponen and M.P. Tarvainen. A robust algorithm for heart rate variability time series artefact correction using novel beat classification. *J Med Eng & Technol*, 43(3):173–181, 2019.
- [15] D.A. Litvack, T.F. Oberlander, L.H. Carney, and J.P. Saul. Time and frequency domain methods for heart rate variability analysis: a methodological comparison. *Psychophysiol*, 32:492–504, 1995.
- [16] M.D. Mifflin, S.T. St Jeor, L.A. Hill, B.J. Scott, S.A. Daugherty, and Y.O. Koh. A new predictive equation for resting energy expenditure in healthy individuals. *J Am Diet Assoc*, 2005.
- [17] I.P. Mitov. A method for assessment and processing of biomedical signals containing trend and periodic components. *Med Eng Phys*, 20(9):660–668, November-December 1998.
- [18] R.H. Morton, J.R. Fitz-Clarke, and E.W. Banister. Modeling human performance in running. *J Appl Physiol*, 69(3):1171–7, 1990.
- [19] J-P. Niskanen, M.P. Tarvainen, P.O. Ranta-aho, and P.A. Karjalainen. Software for advanced HRV analysis. *Comput Meth Programs Biomed*, 76(1):73–81, 2004.

References 41

[20] D. Nunan, G.R.H. Sandercock, and D.A. Brodie. A quantitative systematic review of normal values for short-term heart rate variability in healthy adults. *PACE*, 33:1407–1417, November 2010.

- [21] N-J. Olivasse, P.R. Benchimol-Barbosa, and J. Nadal. Refining the deceleration capacity index in phase-rectified signal averaging to assess physical conditioning level. *J Electrocardiol*, 47(3):306–310, 2014.
- [22] J. Pan and W.J. Tompkins. A real-time QRS detection algorithm. *IEEE Trans Biomed Eng*, 32(3):230–236, March 1985.
- [23] C.-K. Peng, S. Havlin, H.E. Stanley, and A.L. Goldberger. Quantification of scaling exponents and crossover phenomena in nonstationary heartbeat time series. *Chaos*, 5:82–87, 1995.
- [24] T. Penzel, J.W. Kantelhardt, L. Grote, J.-H. Peter, and 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):1143–1151, October 2003.
- [25] S.W. Porges and R.E. Bohrer. The analysis of periodic processes in psychophysiological research. In J.T. Cacioppo and L.G. Tassinary, editors, *Principles of Psychophysiology: Physical Social and Inferential Elements*, pages 708–753. Cambridge University Press, 1990.
- [26] J.A. Richman and J.R. Moorman. Physiological time-series analysis using approximate entropy and sample entropy. *Am J Physiol*, 278:H2039–H2049, 2000.
- [27] M.P. Tarvainen, S.D. Georgiadis, P.O. Ranta-aho, and P.A. Karjalainen. Time-varying analysis of heart rate variability signals with Kalman smoother algorithm. *Physiol Meas*, 27(3):225–239, 2006.
- [28] M.P. Tarvainen, J.-P. Niskanen, J.A. Lippponen, P.O. Ranta-aho, and P.A. Karjalainen. Kubios HRV heart rate variability analysis software. *Comp Meth Programs Biomed*, 113(1):210–220, 2014.
- [29] M.P. Tarvainen, P.O. Ranta-aho, and P.A. Karjalainen. An advanced detrending method with application to HRV analysis. *IEEE Trans Biomed Eng*, 49(2):172–175, February 2002.
- [30] 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):1043–1065, March 1996.
- [31] C.L. Webber Jr. and J.P. Zbilut. Dynamical assessment of physiological systems and states using recurrence plot strategies. *J Appl Physiol*, 76:965–973, 1994.
- [32] E.J.M. Weber, C.M. Molenaar, and M.W. van der Molen. A nonstationarity test for the spectral analysis of physiological time series with an application to respiratory sinus arrhythmia. *Psychophysiol*, 29(1):55–65, January 1992.
- [33] J.P. Zbilut, N. Thomasson, and C.L. Webber. Recurrence quantification analysis as a tool for the nonlinear exploration of nonstationary cardiac signals. *Med Eng Phys*, 24:53–60, 2002.