

ORIGINAL RESEARCH ARTICLE

Rapid assessment of cardiac autonomic modulation and adaptive stress responses: Automatic calculation of time-varying parasympathetic, sympathetic, and Baevsky stress indexes

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Abstract

Cardiac autonomic modulation (CAM), which is regulated by the balance between the sympathetic and parasympathetic nervous systems, is involved in various physiological and pathological conditions. Heart rate variability (HRV) analysis has been used to explore the complex relationship between the brain and heart, as described by Porges' polyvagal theory and Thayer's neurovisceral integration model. Recently, an automated calculation of new parasympathetic, sympathetic, and Baevsky stress indexes based on HRV parameters has been introduced for faster and more comprehensive CAM assessment, though their normal ranges remain undefined. This study aimed to determine the average values of these indexes in a healthy population of different ages during rest, daily activities, non-rapid eye movement sleep, graded physical effort, and acute psychophysiological stress. At rest, the parasympathetic and sympathetic indexes were consistently within the proposed normal range and inversely related. However, Baevsky stress index values from Kubios were higher than expected, conflicting with the assumption that they are simply the square root of those calculated using the original formula. Despite this, time-varying assessment of all indexes can provide valuable insights into CAM adaptation during physical effort and acute psychophysiological stress in real-world critical situations. Notably, our novel finding shows that the inverse correlation between parasympathetic and sympathetic/stress indexes under stress is better explained by non-linear functions, offering a potential new measure of brain–heart interaction during real-life critical events.

Keywords: Heart rate variability; Autonomic nervous system; Sympathetic nervous system; Parasympathetic nervous system; Baevsky stress index; Psychophysiological stress

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1. Introduction

Cardiac autonomic modulation (CAM), regulated by the balance between the sympathetic (SNS) and parasympathetic nervous systems (PNS), is vital in both health and disease, making its evaluation crucial.^{1,2} The study of heart rate variability (HRV), which reflects autonomic nervous system (ANS) function, began in the 1960s, initially for monitoring astronauts in space.³ Since the 1980s, HRV analysis has been widely used as a non-invasive method to evaluate CAM in clinical settings, with autonomic imbalance recognized as a strong predictor of all-cause cardiac mortality.⁴

Early HRV assessments followed a “reductionist” approach,⁵ relying on linear methods such as time-domain (TD) and frequency-domain (FD) analysis.^{4,6,7} Many studies were based on the premise that increased sympathetic activity corresponds with decreased parasympathetic activity, with the spectral low frequency/high frequency (LF/HF) ratio suggested as a marker for stress responses, though this remains debated.^{7,8}

HRV tends to decline with age due to structural and functional changes,⁹⁻¹² and significant reductions in HRV are associated with lower life expectancy and various diseases.^{5,9-18} It is important to note that the LF component is not exclusively a marker of sympathetic activity, as both SNS and PNS influence LF.⁵ In addition, the very-low-frequency (VLF) band may reflect broader systemic stress, not just cardiac stress.¹⁹⁻²¹

A more systematic perspective suggests that HRV is not only a marker of ANS dysfunction but also a means for understanding interactions between bodily systems, especially between the brain and cardiovascular system.⁵ In health, “organized variability” is present, whereas disease leads to a “decomplexification” of this variability, resulting in more periodic and cyclic behaviors, as seen in conditions such as Cheyne–Stokes breathing, Parkinson’s disease tremors, or cyclic neutrophil oscillations in chronic myelogenous leukemia.²²

The vagal system plays a key role in brain–heart interaction, particularly in stress regulation, as explained by Porges’ “polyvagal theory”^{23,24} and Thayer’s “neurovisceral integration model,” which links HRV to emotional and stress responses.²⁵⁻³⁰

To better study these complex interactions, non-linear (NL) mathematical methods have been introduced. For instance, NL entropy is associated with vagal activity, whereas recurrence plot analysis reflects sympathetic activation.^{31,32} In stress-related studies, such as those examination-induced stress, combining linear and NL methods has improved stress detection.³³ Mathematical

models for the time-varying analysis of both linear and NL HRV parameters have enhanced the ability to assess CAM responses to clinical or real-world stressors, even in real time.³⁴⁻³⁹

Recently, the software has been developed to automatically compute time-varying parasympathetic (PNSi), sympathetic (SNSi), and Baeovsky stress (BSTRI) indexes, enabling faster assessments of CAM, particularly in acute stress situations.⁴⁰ Although HRV parameters are well-established, the normal ranges for these new indexes, especially BSTRI, remain unclear, particularly in stressful conditions.⁴¹⁻⁴⁶

This pilot study aims to determine the average values of PNSi, SNSi, and BSTRI in a healthy population during various activities (rest, daily activities, non-rapid eye movement (NREM) sleep, graded physical effort, and acute psychophysiological stress) to create a preliminary reference for CAM assessment in real-world high-stress or pathological conditions, such as dysautonomic syndromes.

2. Methods

2.1. Study population, ethics, and inclusion criteria

We retrospectively analyzed 24-h Holter electrocardiogram (ECG) recordings from 104 healthy individuals, categorized into three age groups: Age1 (16–34 years), Age2 (35–52 years), and Age3 (53–84 years), all consecutively evaluated in our clinic for fitness or clinical evaluations. Among these participants, 16 were police officers monitored during fitness assessments and highly stressful, realistic tactical training. In addition, ECG data from a 75-year-old healthy male who volunteered to be monitored 3 times/week from April to November 2022 during training on a professional bicycle ergometer (Technogym, Italy) were included (Table 1).

All participants were free of cardiovascular and neurological conditions based on clinical history, physical examinations, and laboratory results. They were required to refrain from smoking, consuming coffee, or using any substances or medications that could affect their natural sympathovagal balance for at least 24 h before ECG recording.

All participants provided informed consent for ECG recording and monitoring, primarily related to preventive checkups for non-competitive sports or fitness-for-duty evaluations. They also agreed to the anonymous use of their ECG data for scientific and publication purposes. All recording procedures were conducted in accordance with good clinical practice (GCP) guidelines and ethical standards outlined in the Helsinki Declaration of 1975, revised in 2013.⁴⁷

2.2. HRV recordings and analysis

We conducted 24-h 12-lead Holter ECG recordings using the H12 (Mortara Instruments). Real-time ECG monitoring during tactical and physical training was carried out with a 3-lead wearable ECG (Nuubo, Spain).

For longitudinal monitoring during physical training, each session included 10 min of baseline rest, 5 min of warm-up, 30 min of exercise, and 10 min of recovery. The training workload was categorized by age into low (65–75 watts/min), moderate (75–85 watts/min), semi-intense (85–95 watts/min), and intense (95–110 watts/min). For this study, ECG data from 40 training sessions (10 sessions for each workload) were analyzed.

HRV parameters were calculated using Kubios Premium software (version 3.5.0) in TD, FD, and NL methods, along with time-varying algorithms,⁴⁰ following “detrending” with the “smooth priors” function (lambda = 500).

Short-term HRV was calculated over 5 min⁴ during regular daily activities and NREM sleep for all participants, as well as during highly stressful police tactical training (16 cases). In addition, HRV parameters were assessed from 2-min intervals, as illustrated in Figure 1, to evaluate

the reliability of PNSi, SNSi, and BSTRi measurements from these shorter intervals.

For the training sessions, HRV parameters were calculated using 2-min intervals taken at baseline rest, every 4 min during exertion, and at the first, fifth, and tenth min of recovery. ECG data from 40 training sessions (10 sessions for each workload) were analyzed. The PNSi, SNSi, and BSTRi values resulting from each workload were averaged and compared to evaluate reproducibility across sessions with the same workload.

Kubios calculates the PNSi by integrating three parameters: the mean RR interval, the RMSSD (the mean square root of successive RR interval differences, closely related to parasympathetic cardiac activation), and the SD1 index from Poincaré plot (in normalized units), which correlates with the RMSSD. The PNS index is considered normal during rest if it falls within ±2 standard deviations (SD) of the normal population distribution.⁴⁰

The SNSi is computed using three parameters: the mean HR interval, the BSTRi, and the SD2 index from the Poincaré plot (in normalized units), which correlates with SDNN.⁴² Similar to the PNSi, the SNS index is deemed normal if it is within ±2 SD of the normal population distribution. During psychophysiological stress or high-intensity exercise, significantly lower PNSi and higher SNSi values are expected.

The BSTRi is a geometric measure of HRV that indicates stress on the cardiovascular system, calculated using the following formula:

$$SI = \frac{AMo}{Mo} MxDMn \tag{I}$$

Table 1. Demographics of the studied participants

Subgroups	No. of cases	Sex		Age range (years)
		Male	Female	
Age1	36	10	26	16–34
Age2	29	15	14	35–52
Age3	23	10	13	53–84
Police officers	16	15	1	31–50
Senior training	1	1		75

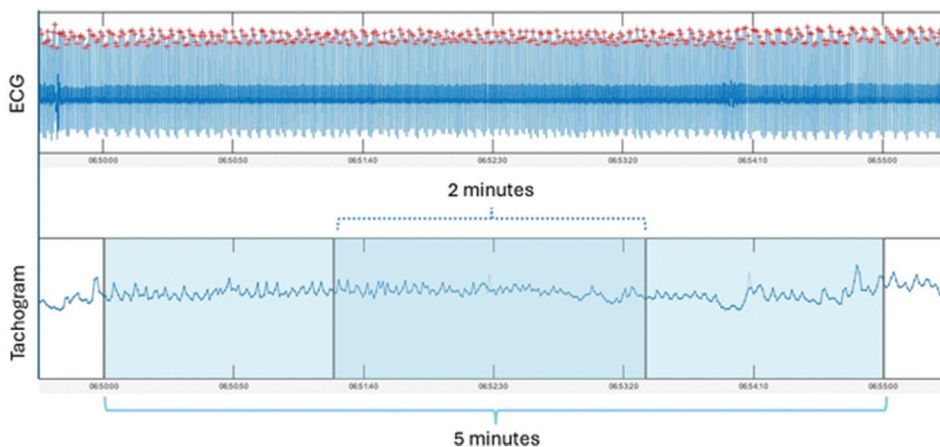


Figure 1. Time segment selection method. The upper row shows 6 min of ECG recording, whereas the lower row displays examples of 5-min (light sky blue) and 2-min (darker sky blue) tachogram segments selected. Abbreviation: ECG: Electrocardiogram.

where AMo is the mode amplitude (the percentage of intervals corresponding to the mode value relative to the sample size), Mo is the mode, and MxDm represents the degree of interval variability, calculated from the difference between the maximum (Mx) and minimum (Mn) intervals. According to Baevsky, BSTRi values are considered normal at rest when they range from 80 to 150 conventional units and are 1.5–2 times higher during physical or emotional stress. High BSTRi values suggest reduced variability and increased sympathetic cardiac activation.^{43,44} In Kubios, BSTRi values are expressed as the square root of the original Baevsky values.⁴⁰

To calculate both PNSi and SNSi, each parameter is first compared to the normal population values reported by Nunan.⁴¹ These values are then scaled using the SD of the normal population and refined with undisclosed proprietary weighting factors to ensure robust and reliable index values. This process considers the relationships between exercise intensity, heart rate, and HRV.⁴⁰

2.3. Statistical analysis

Data were input into SPSS (21.0 version, Chicago, Illinois) for quantitative analysis. All data were examined for expected ranges, outliers, and abnormal values. Continuous variables are presented as mean and SD. Differences between groups were assessed using either parametric or non-parametric tests, as appropriate. A $P < 0.05$ was

considered statistically significant. The Pearson correlation was used to evaluate the relationships between selected quantitative variables, with a correlation considered strong if $R > 0.7$.

3. Results

3.1. Population

The demographics of the 105 healthy participants are summarized in Table 1.

3.2. Linear and NL HRV parameters

3.2.1. Variations in HRV parameters based on short-term interval duration

Compared to those calculated from 5-min intervals, the average values of most HRV parameters were not significantly different when calculated from 2-min intervals across all examined situations (daily activity, NREM sleep, and acute psychophysiological stress). Significant differences ($P < 0.05$) for 5-min intervals were found only for certain recurrence plot parameters (in all conditions), approximated entropy (not under stress), the triangular index, TINN (during NREM sleep), and minimum heart rate under stress (Table 2).

For clarity, only a selection of Kubios-calculated parameters will be presented in the following tables.

Table 2. Difference in each HRV parameter calculated from short-term time segments of 2 and 5 min, during regular daily activity (104 cases), NREM sleep (104 cases), and high-stress tactical training (16 police officers)

	Daily activity				NREM sleep				High stress								
	2 min interval		5 min		n	P	2 min interval		5 min interval		n	P	2 min interval		5 min interval		
	Mean	SD	Mean	SD			Mean	SD	Mean	SD			Mean	SD	Mean	SD	
MeanRR <i>ms</i>	734.3	120.1	731.9	116.8	104		977.8	148.7	975.0	154.0	104		438.0	75.0	445.3	70.2	16
SDNN <i>ms</i>	31.7	15.1	31.2	13.7	104		27.2	12.5	27.5	12.2	104		13.7	13.1	15.0	12.6	16
MeanHR <i>bpm</i>	83.8	13.2	84.0	13.0	104		62.7	9.1	63.0	9.9	104		140.2	20.2	137.3	17.7	16
MinHR <i>bpm</i>	74.7	11.5	72.7	10.9	104		59.6	8.6	59.1	9.2	104		122.9	20.9	106.6	16.8	16 #
MaxHR <i>bpm</i>	93.7	14.5	97.0	14.9	104		65.7	9.7	67.4	11.6	104		155.6	18.6	160.1	17.7	16
RMSSD <i>ms</i>	22.3	12.1	21.8	11.0	104		32.7	16.0	32.7	16.1	104		6.4	6.1	7.0	6.5	16
pNN50	5.1	8.5	4.6	7.7	104		14.1	16.5	14.2	16.5	104		0.4	1.2	0.6	1.8	16
HRVtriang index	7.6	2.6	8.3	3.0	104		7.1	2.4	7.8	2.8	104 #		3.6	2.3	3.5	2.5	16
TINN <i>ms</i>	140.9	65.0	155.9	67.7	104		117.3	52.2	135.4	62.2	104 #		63.9	57.2	84.3	59.7	16
DC <i>ms</i>	18.3	12.4	17.6	11.5	104		28.0	18.6	27.9	17.9	104		5.4	9.6	5.2	8.6	16
DCmod <i>ms</i>	23.1	12.3	22.5	11.4	104		36.8	18.6	36.7	18.6	104		6.5	7.4	6.8	8.0	16
AC <i>ms</i>	-18.1	12.7	-17.5	11.9	104		-28.9	18.3	-28.9	18.1	104		-4.2	6.0	-4.3	5.8	16
ACmod <i>ms</i>	-23.7	14.0	-23.0	12.8	104		-38.7	19.9	-38.6	20.0	104		-5.8	5.6	-6.3	6.6	16
VLFpov FFT <i>ms</i> ²	98.6	130.7	99.4	102.8	104		39.0	60.4	45.5	58.3	104		25.0	29.3	25.7	27.0	16
LFpov FFT <i>ms</i> ²	880.4	1,051.9	844.5	921.3	104		250.1	364.3	286.3	397.6	104		276.6	504.1	317.2	596.6	16

(Contd...)

Table 2. (Continued)

	Daily activity					NREM sleep					High stress							
	2 min interval		5 min		<i>n</i>	<i>P</i>	2 min interval		5 min interval		<i>n</i>	<i>P</i>	2 min interval		5 min interval		<i>n</i>	<i>P</i>
	Mean	SD	Mean	SD			Mean	SD	Mean	SD			Mean	SD	Mean	SD		
HFpow FFT <i>ms</i> ²	218.0	336.6	203.2	235.3	104	508.7	647.1	512.6	662.8	104	47.8	81.6	37.6	62.2	16			
VLFpow FFT log	3.9	1.3	4.1	1.1	104	3.1	1.0	3.3	0.9	104	2.5	1.3	2.7	1.2	16			
LFpow FFT log	6.2	1.1	6.3	1.0	104	5.1	0.9	5.3	0.8	104	4.1	1.9	4.3	1.8	16			
HFpow FFT log	4.8	1.1	4.8	1.1	104	5.8	0.9	5.8	0.9	104	2.3	1.9	2.4	1.7	16			
VLFpow FFT	9.4	6.5	10.2	6.5	104	6.0	4.7	6.4	4.9	104	18.0	11.7	18.0	13.6	16			
LFpow FFT	72.7	10.5	71.9	10.0	104	32.5	12.1	35.1	12.0	104	70.3	8.8	71.3	11.8	16			
HFpow FFT	17.9	8.5	17.8	7.8	104	61.5	14.1	58.5	14.2	104	11.7	6.8	10.7	5.3	16			
LFpow FFT nu	80.1	9.4	80.0	8.9	104	34.8	13.6	37.8	14.0	104	86.3	7.3	87.2	5.8	16			
HFpow FFT nu	19.9	9.4	19.9	8.9	104	65.1	13.6	62.2	14.0	104	13.7	7.3	12.8	5.8	16			
TOTpow FFT <i>ms</i> ²	1,197.2	1,342.9	1,147.4	1,168.6	104	798.1	955.8	844.7	1,014.2	104	349.4	605.0	380.5	681.8	16			
LF/HFratio FFT	5.2	3.3	5.1	3.0	104	0.6	0.7	0.8	1.2	104	9.9	9.2	8.7	4.9	16			
VLFpow AR <i>ms</i> ²	121.9	127.8	121.7	106.8	104	51.0	56.6	55.8	50.0	104	41.4	66.2	48.9	69.5	16			
LFpow AR <i>ms</i> ²	820.0	907.9	783.4	767.1	104	265.4	395.7	279.2	340.9	104	270.6	506.6	317.6	588.5	16			
HFpow AR <i>ms</i> ²	210.3	293.9	201.0	248.7	104	488.9	636.9	497.5	635.5	104	30.9	54.1	33.6	56.4	16			
VLFpow AR log	4.4	1.0	4.5	0.9	104	3.6	0.8	3.7	0.7	104	2.8	1.5	3.2	1.3	16			
LFpow AR log	6.2	1.1	6.2	1.0	104	5.1	0.9	5.2	0.9	104	4.1	1.9	4.6	1.5	16			
HFpow AR log	4.7	1.1	4.8	1.1	104	5.7	0.9	5.7	0.9	104	2.2	1.6	2.5	1.5	16			
VLFpow AR	12.1	5.1	12.8	5.0	104	7.9	4.8	8.3	4.7	104	21.1	11.0	18.3	8.7	16			
LFpow AR	70.5	10.0	69.9	9.3	104	33.3	11.4	35.0	12.4	104	68.4	9.6	72.5	8.1	16			
HFpow AR	17.4	8.5	17.3	7.6	104	58.8	13.8	56.7	14.8	104	10.5	4.9	9.2	4.4	16			
LFpow AR nu	80.1	9.7	80.0	9.0	104	36.4	13.4	38.5	14.7	104	86.9	5.7	88.9	5.0	16			
HFpow AR nu	19.8	9.7	19.9	8.9	104	63.5	13.4	61.5	14.7	104	13.1	5.7	11.1	5.0	16			
TOTpow AR <i>ms</i> ²	1,152.5	1,204.8	1,106.4	1,065.9	104	805.6	1,003.0	832.7	944.4	104	342.9	614.0	400.0	703.9	16			
LF/HF ratio AR	5.1	2.8	5.1	2.8	104	0.7	0.7	0.8	1.1	104	8.0	3.7	9.3	3.4	16			
RESP Hz	0.2	0.1	0.2	0.1	104	0.3	0.0	0.3	0.1	104	0.4	0.1	0.5	0.1	16			
SD1 <i>ms</i>	15.8	8.6	15.4	7.8	104	23.2	11.4	23.1	11.4	104	4.5	4.3	5.0	4.6	16			
SD2 <i>ms</i>	41.9	19.9	41.1	18.1	104	30.4	14.1	31.1	13.7	104	18.8	18.0	20.6	17.2	16			
SD2/SD1 ratio	2.7	0.7	2.8	0.7	104	1.4	0.3	1.4	0.4	104	4.1	1.3	4.3	1.1	16			
ApEn	0.8	0.1	1.2	0.1	104 #	0.6	0.1	1.1	0.1	104 #	1.0	0.2	1.0	0.2	16			
SampEn	1.5	0.3	1.5	0.2	104	1.9	0.4	1.9	0.2	104	1.2	0.4	1.0	0.3	16			
D2	1.3	1.2	1.2	1.3	104	1.2	1.3	1.2	1.4	104	0.3	0.7	0.3	0.7	16			
DFA1	1.4	0.2	1.4	0.2	104	0.7	0.2	0.8	0.2	104	1.5	0.2	1.6	0.1	16			
DFA2	0.5	0.1	0.5	0.1	104	0.3	0.1	0.3	0.1	104	0.9	0.3	0.8	0.3	16			
RP_Lmean beats	9.5	2.7	10.3	2.6	104 #	7.7	2.2	8.5	2.2	104 #	14.0	2.2	20.7	5.6	16 #			
RP_Lmax beats	107.3	45.6	204.5	108.9	104 #	38.1	16.2	63.5	31.2	104 #	205.0	59.3	516.4	185.6	16 #			
RP_REC	29.1	7.6	30.6	6.9	104	17.5	4.9	18.9	5.0	104 #	40.4	4.8	48.8	8.1	16 #			
RP_DET	97.6	1.8	98.0	1.3	104	94.8	2.4	95.6	1.8	104 #	99.1	0.9	99.5	0.3	16			
RP_ShanEn	2.9	0.3	3.1	0.3	104 #	2.6	0.3	2.8	0.3	104 #	3.4	0.2	3.8	0.3	16 #			

Note: All parameters calculated by Kubios are included, with statistically significant differences ($P < 0.05$) among groups indicated by # in the *P* columns. Abbreviations: min: Minute; *bpm*: Beats per minute; *ms*: Milliseconds.

3.2.2. Variations in selected HRV parameters related to daily activity and NREM sleep

As expected, the average values of most HRV parameters were significantly different between daily activity and NREM sleep, with no significant effect from the duration of the selected short-term intervals (Table 3).

3.2.3. Age-related differences in HRV parameters

Consistent with previous studies, a significant decline in HRV was observed in most TD and FD parameters associated with aging, during both daily activity and NREM sleep. Statistically significant differences were noted between the Age3 values (C in Table 4, which shows

Table 3. Variations of selected HRV parameters during daily activity and NREM sleep conditions (values calculated from 2-min and 5-min time segments are provided)

	2-min interval					P	5-min interval					P
	Daily activity		NREM sleep				Daily activity		NREM sleep			
	Mean	SD	Mean	SD	n		Mean	SD	Mean	SD	n	
MeanRR <i>ms</i>	734.26	120.12	977.81	148.69	104	#	731.95	116.82	975.00	153.99	104	#
SDNN <i>ms</i>	31.74	15.06	27.21	12.53	104		31.15	13.69	27.53	12.23	104	
MeanHR <i>bpm</i>	83.78	13.20	62.71	9.07	104	#	83.97	13.00	63.05	9.93	104	#
MinHR <i>bpm</i>	74.73	11.50	59.63	8.58	104	#	72.70	10.95	59.15	9.18	104	#
MaxHR <i>bpm</i>	93.66	14.48	65.65	9.70	104		97.02	14.85	67.36	11.63	104	#
RMSSD <i>ms</i>	22.30	12.11	32.71	15.99	104	#	21.77	11.01	32.65	16.10	104	#
pNN50	5.07	8.49	14.08	16.49	104	#	4.63	7.65	14.21	16.50	104	#
HRVtriang index	7.56	2.61	7.09	2.43	104		8.27	3.04	7.83	2.77	104	
TINN <i>ms</i>	140.87	64.96	117.31	52.15	104		155.89	67.74	135.38	62.20	104	
VLFpow FFT <i>ms</i> ²	98.64	130.69	39.03	60.39	104		99.42	102.79	45.55	58.30	104	#
LFpow FFT <i>ms</i> ²	880.39	1,051.88	250.05	364.28	104	#	844.50	921.31	286.27	397.62	104	#
HFpow FFT <i>ms</i> ²	217.98	336.56	508.72	647.12	104		203.24	235.34	512.60	662.76	104	#
LFpow FFT nu	80.09	9.42	34.84	13.61	104	#	80.04	8.93	37.78	13.98	104	#
HFpow FFT nu	19.88	9.41	65.09	13.61	104	#	19.93	8.92	62.17	13.98	104	#
TOTpow FFT <i>ms</i> ²	1,197.25	1,342.90	798.11	955.76	104		1,147.42	1,168.60	844.74	1,014.20	104	
LF/HF ratio FFT	5.21	3.31	0.65	0.66	104	#	5.11	3.01	0.81	1.22	104	#
SD1 <i>ms</i>	15.82	8.60	23.23	11.36	104	#	15.41	7.80	23.13	11.40	104	#
SD2 <i>ms</i>	41.85	19.89	30.42	14.12	104	#	41.12	18.08	31.06	13.66	104	#
SD2/SD1 ratio	2.75	0.67	1.36	0.30	104	#	2.77	0.67	1.42	0.43	104	#
ApEn	0.83	0.09	0.65	0.09	104	#	1.16	0.09	1.08	0.09	104	#
SampEn	1.54	0.30	1.95	0.39	104	#	1.53	0.25	1.89	0.22	104	#
DFA1	1.37	0.21	0.74	0.19	104	#	1.38	0.21	0.77	0.20	104	#
DFA ²	0.46	0.15	0.29	0.13	104	#	0.48	0.13	0.30	0.11	104	#
RP_Lmean <i>beats</i>	9.51	2.71	7.70	2.16	104	#	10.28	2.62	8.48	2.21	104	#
RP_Lmax <i>beats</i>	107.28	45.60	38.12	16.23	104	#	204.55	108.91	63.53	31.21	104	#
RP_REC	29.06	7.63	17.48	4.94	104	#	30.59	6.92	18.91	5.05	104	#
RP_DET	97.62	1.77	94.79	2.38	104	#	97.96	1.32	95.55	1.81	104	#
RP_ShanEn	2.91	0.29	2.60	0.29	104	#	3.09	0.28	2.84	0.29	104	#
RESP Hz	0.24	0.09	0.26	0.04	104		0.25	0.09	0.27	0.05	104	

Note: Statistically significant differences ($P < 0.05$) among groups are marked with # in the P-value columns.

Abbreviations: NREM: Non-rapid eye movement; RR: Interval between 2 subsequent R peaks; SDNN: Standard deviation Normal to Normal; RMSSD: Root Mean Square of Successive Differences; pNN50: The percentage of successive normal cardiac interbeat intervals > 50 msec HR heart rate; HF: High frequency; LF: Low frequency; TOT: total. pwr: power; SD1 and SD2: Poincare Plot parameters; DFA1 and DFA2: Detrended Fluctuation Analysis alfa parameters; ApEn: Approximated Entropy; SamEn: Sample entropy; RP: Recurrence Plot parameters.

Table 4. TD, FD, and NL parameters calculated during daily activity and NREM sleep across the three age groups.

	Daily activity						(A) (B) (C)			NREM sleep						(A) (B) (C)		
	(A) Age1 (n. 37)		(B) Age2 (n. 44)		(C) Age3 (n. 23)		<i>P</i> <0.05			(A) Age1 (n. 37)		(B) Age2 (n. 44)		(C) Age3 (n. 23)		<i>P</i> <0.05		
	Mean	SD	Mean	SD	Mean	SD				Mean	SD	Mean	SD	Mean	SD			
MeanRR <i>ms</i>	690.1	96.1	737.8	113.8	788.0	131.0		A	917.9	151.2	992.3	149.7	1033.8	141.9			A	
SDNN <i>ms</i>	34.5	14.6	33.0	12.9	22.2	9.7	C	C	28.5	9.7	31.5	14.3	18.3	5.2	C	C		
MeanHR <i>bpm</i>	88.7	13.1	83.1	12.2	78.0	11.8	C		66.8	9.0	62.0	10.5	59.1	8.6	C			
MinHR <i>bpm</i>	75.3	10.7	71.6	10.7	70.7	11.5			62.0	8.8	58.1	9.6	56.7	8.2				
MaxHR <i>bpm</i>	104.0	13.4	96.0	14.4	87.7	12.5	B	C	71.9	10.3	66.3	12.5	62.0	9.4	C			
RMSSD <i>ms</i>	23.5	12.9	22.6	10.7	17.4	6.8			33.6	13.1	37.4	19.3	22.0	5.9	C	C		
pNN50	6.0	9.8	5.1	7.1	1.5	2.3			15.1	15.4	19.4	18.8	2.9	3.1	C	C		
HRVtriang index	9.2	3.6	8.5	2.5	6.3	2.2	C	C	8.3	2.3	8.7	3.0	5.5	1.4	C	C		
TINN <i>ms</i>	172.3	72.4	164.9	62.4	112.2	52.0	C	C	141.8	52.2	153.9	70.1	89.7	33.4	C	C		
VLFpww FFT <i>ms</i> ²	101.3	77.9	107.9	129.0	80.2	80.3			42.8	62.3	60.1	65.5	22.1	13.0		C		
LFpww FFT <i>ms</i> ²	919.5	820.9	1,017.3	1,118.3	393.2	380.2		C	272.9	200.8	393.2	559.0	103.2	72.5		C		
HFpww FFT <i>ms</i> ²	281.6	323.0	196.7	164.6	89.6	108.2	C		496.1	432.2	699.7	891.6	181.2	109.0		C		
LFpww FFT nu	78.2	8.2	81.5	9.8	80.3	8.1			37.7	12.6	38.5	16.9	36.4	9.8				
HFpww FFT nu	21.8	8.2	18.5	9.8	19.6	8.1			62.2	12.6	61.4	16.9	63.5	9.8				
TOTpww FFT <i>ms</i> ²	1,302.8	1,146.4	1,322.2	1,341.0	563.1	519.5	C	C	812.1	593.8	1,153.4	1,378.5	306.7	172.5		C		
LF/HF ratio FFT	4.5	3.3	5.7	3.0	4.9	2.3			0.7	0.7	1.0	1.7	0.6	0.3				
SD1 <i>ms</i>	16.7	9.2	16.0	7.5	12.3	4.8			23.8	9.3	26.5	13.7	15.6	4.2	C	C		
SD2 <i>ms</i>	45.8	18.8	43.7	16.9	28.8	13.4	C	C	32.3	10.7	35.5	15.8	20.6	6.4	C	C		
SD2/SD1 ratio	2.9	0.6	2.9	0.6	2.3	0.7	C	C	1.4	0.3	1.5	0.6	1.3	0.3				
ApEn	1.2	0.1	1.1	0.1	1.2	0.1			1.1	0.1	1.1	0.1	1.1	0.1				
SampEn	1.5	0.3	1.5	0.2	1.7	0.2		A B	1.9	0.2	1.8	0.2	2.0	0.2		B		
D2	1.4	1.4	1.4	1.2	0.5	0.9	C	C	1.4	1.5	1.7	1.4	0.1	0.2	C	C		
DFA1	1.4	0.2	1.4	0.2	1.3	0.3		C	0.8	0.2	0.8	0.2	0.8	0.2				
DFA2	0.5	0.1	0.4	0.1	0.5	0.1		B	0.3	0.1	0.3	0.1	0.3	0.1				
RP_Lmean <i>beats</i>	10.5	3.0	10.5	2.2	9.4	2.6			8.4	2.0	9.0	2.3	7.5	2.0		C		
RP_Lmax <i>beats</i>	229.8	114.7	209.1	107.5	155.3	88.0	C		64.0	29.4	67.6	34.4	55.1	26.9				
RP_REC	31.3	7.5	31.4	6.0	27.9	7.2			18.8	3.9	19.8	5.8	17.3	5.0				
RP_DET	98.2	1.1	98.2	1.2	97.2	1.7	C	C	95.7	1.5	96.0	1.8	94.5	2.1	C	C		
RP_ShanEn	3.1	0.3	3.1	0.2	3.0	0.3			2.8	0.3	2.9	0.3	2.7	0.3		C		
RESP <i>Hz</i>	0.2	0.1	0.3	0.1	0.3	0.1		A	0.3	0.0	0.3	0.1	0.3	0.0				

Note: Statistically significant differences among groups are indicated by A, B, and C in the *P*<0.05 columns.

Abbreviations: TD: Time domain; FD: Frequency domain; NL: Non-linear; RR: The interval between 2 subsequent R peaks; SDNN: Standard deviation Normal to Normal; RMSSD: Root Mean Square of Successive Differences; HR: heart rate; HF: High Frequency; LF: Low Frequency; TOT: total; pwr: Power; SD1 and SD2: Poincare Plot parameters; DFA1 and DFA2: Detrended Fluctuation Analysis alfa parameters; ApEn: Approximated Entropy; SampEn: Sample entropy; RP: Recurrence Plot parameters.

data analyzed from 5-min intervals) and those of both Age1 and Age2 groups (A and B in Table 4).

During daily activity, sympathetic modulation was indicated by higher LFpower (919.5 msec² in Age1, 1017.3 msec² in Age2, and 393.0 msec² in Age3) and LF/HF ratio (4.5 in Age1, 5.7 in Age2, and 4.9 in Age3), along with

lower RMSSD (23.5 ms in Age1, 22.6 in Age2, and 17.4 in Age3), HFpower (281.6 msec² in Age1, 196.7 msec² in Age2, and 89.6 msec² in Age3), and SD1 (16.7 ms in Age1, 16.0 in Age2, and 12.3 in Age3).

In addition, the VLFpower values were higher during activity (101.3 msec² in Age1, 107.9 msec² in Age2, and

80.2 msec² in Age3) compared to NREM sleep (42.8 msec² in Age2, 60.1 msec² in Age2, and 22.1 msec² in Age3), with a statistically significant difference noted only between Age2 and Age3 during NREM (Table 4).

3.3. Kubios indexes

The PNSi, SNSi, and BSTRi values showed no significant differences when calculated from 2- and 5-min intervals during daily activity and NREM sleep (Table 5).

3.3.1. Age-related variations in Kubios indexes during daily activity and NREM sleep

Average PNSi values fell within the recommended normal range,⁴⁰ with no significant age-related variations noted during either daily activity or NREM sleep.

Average SNSi values were slightly above the normal range during daily activity but remained within normal limits during NREM sleep, particularly in the Age2 group (0.36 ± 1.33) compared to Age1 (0.82 ± 1.17) and Age3 (0.92 ± 0.98).

Average BSTRi values were considerably lower than the “normal range” indicated in the original literature,^{43,44} as Kubios’ BSTRi is calculated as the square root of Baevsky’s values.⁴⁰ There were no significant differences within each age group between daily activity and NREM sleep. However, a gradual increase was observed from Age1 to Age3, with Age3 values significantly higher (*P* < 0.05) than

those of Age1 and Age2 during both daily activity and NREM sleep (Table 6).

3.3.2. Kubios indexes during physical training

Figure 2 illustrates an example of Kubios’ synthetic output for PNSi and SNSi during regular daily activity and physical effort.

Average values for PNSi, SNSi, and BSTRi were significantly different (*P* < 0.05) between measurements taken at rest and at peak effort, with PNSi decreasing and SNSi and BSTRi increasing. This trend reversed at 1 and 5 min into recovery (Table 7X) and also varied across different training sessions (Table 7Y).

As expected, the relationship between SNSi and BSTRi was strictly linear (*R*² = 0.98), whereas the inverse

Table 5. Average values of PNSi, SNSi, and BSTRi for all 104 participants during daily activity and NREM sleep, calculated from 2- and 5-min time segments

	Daily activity				NREM sleep					
	2-min interval		<i>P</i>	5-min interval		2-min interval		<i>P</i>	5-min interval	
	Mean	SD		Mean	SD	Mean	SD	Mean	SD	
PNSi	-1.46	0.80	<i>ns</i>	-1.49	0.76	0.12	0.92	<i>ns</i>	0.09	0.95
SNSi	2.47	1.74	<i>ns</i>	2.30	1.64	0.77	1.19	<i>ns</i>	0.65	1.22
BSTRi	17.21	6.90	<i>ns</i>	16.01	6.40	16.34	5.12	<i>ns</i>	15.33	4.86

Abbreviations: min: Minute; *ns*: Non-significant.

Table 6. Age-dependent behavior of the parasympathetic (PNSi), sympathetic (SNSi), and Baevsky’s stress (BSTRi) indexes during daily activity and NREM sleep

	Daily activity						NREM sleep									
	(A) Age1 (n. 37)		<i>P</i>	(B) Age2 (n. 44)		<i>P</i>	(A) Age1 (n. 37)		<i>P</i>	(B) Age2 (n. 44)		<i>P</i>	(C) Age3 (n. 23)			
	Mean	SD		Mean	SD		Mean	SD		Mean	SD		Mean	SD		
PNSi	-1.66	0.77	<i>ns</i>	-1.44	0.78	<i>ns</i>	-1.30	0.67	-0.14	0.90	<i>ns</i>	0.29	1.07	<i>ns</i>	0.09	0.71
SNSi	2.44	1.54	<i>ns</i>	2.13	1.64	<i>ns</i>	2.38	1.81	0.82	1.17	<i>ns</i>	0.36	1.33	<i>ns</i>	0.92	0.98
BSTRi	14.77	4.85	<i>ns</i>	15.29	6.31	#	19.39	7.73	14.92	4.50	<i>ns</i>	13.88	4.87	#	18.79	3.77

Note: Statistically significant differences (*P*<0.05) among groups are marked with # in the *P*-value columns.

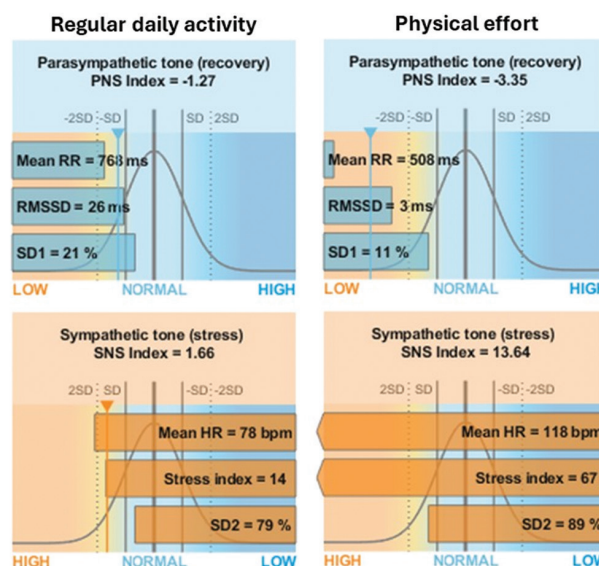


Figure 2. Example of the Kubios⁴⁰ graphic display of PNSi and SNSi, calculated from selected short-term time segments during regular daily activity and physical effort

Abbreviations: PNS: Parasympathetic nervous system; SNS: Sympathetic nervous system; RMSSD: Root mean square of successive differences; HR: Heart rate; SD1 and SD2: Poincare plot parameters.

correlations between PNSi and both SNSi and BSTRi were better fitted by quadratic or cubic functions ($R^2 = 0.92$ for quadratic and $R^2 = 0.96$ for cubic, compared to $R^2 = 0.77$

for linear) and ($R^2 = 0.79$ for quadratic and $R^2 = 0.84$ for cubic, compared to $R^2 = 0.66$ for linear), respectively, when evaluated during physical effort (Figure 3A).

Table 7. Average values of PNSi, SNSi, and BSTRi at rest, at peak effort, and at the first and fifth min of recovery (X). Average values of PNSi, SNSi, and BSTRi across training sessions with different workloads (Y)

X	Selected training phases							
	(A) Rest		(B) Peak effort		(C) First min rec		(D) Fifth min rec	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
PNSi	-1.10	0.46	-3.53	0.40	-2.99	0.55	-2.10	0.27
SNSi	1.92	0.78	17.59	6.10	8.97	4.67	4.47	1.27
BSTRi	18.22	3.54	82.91	26.97	41.30	20.82	26.58	6.43
$P < 0.05$	B C D		A C D		B		B C	
			A C D		A D		A	
			A C D		A D			
Y	Session training workloads (watts/min)							
	(A) 65-75		(B) 75-85		(C) 85-95		(D) 95-110	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
PNSi	-2.21	0.58	-2.44	0.85	-2.53	0.76	-2.73	0.80
SNSi	5.28	2.73	7.26	5.20	7.56	5.36	9.21	6.37
BSTRi	30.34	12.10	38.32	22.15	38.92	22.72	45.39	27.59
$P < 0.05$	C D		D		A		A B C	

Note: Statistically significant differences ($P < 0.05$) among groups are indicated by A, B, C, and D, as applicable. Abbreviation: rec: recovery.

Figure 4 provides an example of the time-varying behavior of PNSi, SNSi, and BSTRi throughout a training session, highlighting four short intervals at baseline rest, medium and peak effort, and during recovery.

3.3.3. Kubios indexes during police tactical training (psychophysiological stress)

The average baseline values of PNSi and SNSi for the 16 police officers were within the suggested normal range,⁴⁰ showing no significant differences when calculated from 2- and 5-min intervals. Average baseline BSTRi values were already above the normal limit before the tactical training began and increased up to 3 times during two subsequent scenarios designed to escalate challenging operational tasks. BSTRi values during stress were higher when calculated from 2-min intervals, though this difference was not statistically significant (Table 8). As expected, the increase in psychophysiological stress from realistic tactical training resulted in a significant increase in SNSi, reaching up to 6 times the upper normal limit, alongside a progressive decrease in PNSi (from -1.4 ± 0.8 to -3.8 ± 0.7) (Table 9).

An example of the time-varying behavior of PNSi and SNSi during a realistic training session is shown in Figure 5A, alongside HRV spectral components (Figure 5B). Individual BSTRi values exceeded 80 conventional units at peak stress (Figure 5C).

Similar to findings during physical training, the relationship between SNSi and BSTRi was strictly linear

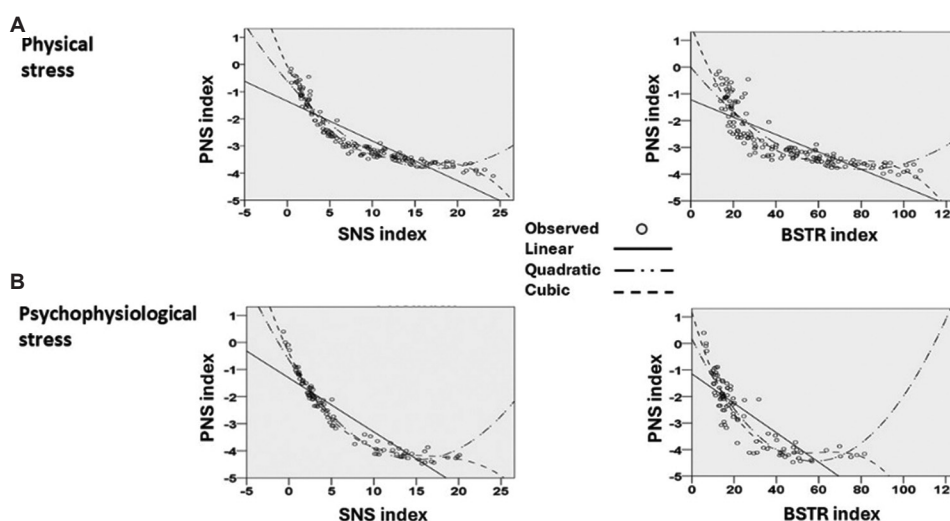


Figure 3. Curves depicting the relationships between PNSi and SNSi (A) and between PNSi and BSTRi (B), illustrating different patterns under physical and psychophysiological stress (see text for details)

Abbreviations: PNS: Parasympathetic nervous system; SNS: Sympathetic nervous system; BSTR: Baevski stress.

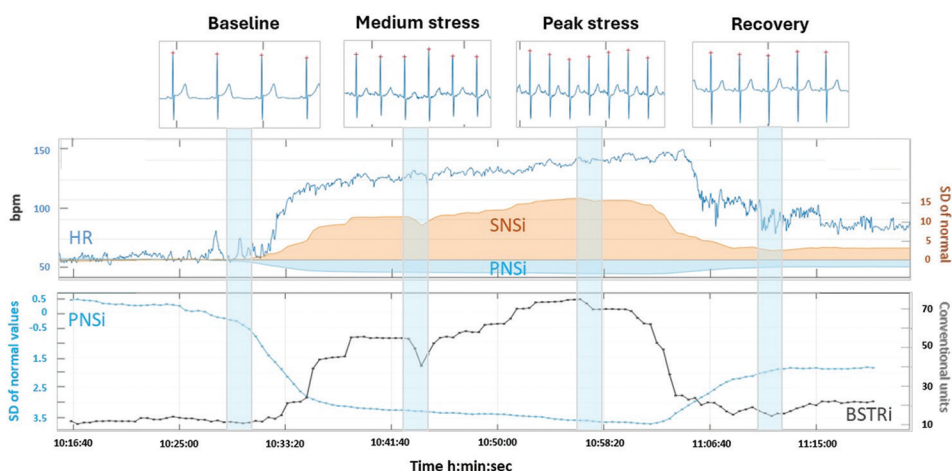


Figure 4. Example of the time-varying behavior of heart rate (HR), PNSi, SNSi, and BSTRi during a training session
 Abbreviations: PNSi: Parasympathetic nervous system index; SNSi: Sympathetic Nervous System index; BSTRi: Baevski stress index; HR: heart rate.

Table 8. Average values of PNSi, SNSi, and BSTRi for the 16 police officers, calculated from 2- and 5-min intervals, before and during realistic tactical scenarios involving varying levels of psychophysiological stress

	Baseline				Medium stress				High stress						
	2-min interval		P	5-min interval		2-min interval		P	5-min interval		2-min interval		P	5-min interval	
	Mean	SD		Mean	SD	Mean	SD		Mean	SD	Mean	SD		Mean	SD
PNSi	-1.4	0.8	ns	-1.6	0.7	-2.8	1.1	ns	-2.8	1.0	-3.8	0.7	ns	-3.7	0.7
SNSi	1.9	1.5	ns	2.1	1.4	7.6	6.4	ns	6.5	5.3	12.5	5.6	ns	10.0	3.7
BSTRi	14.0	5.9	ns	13.9	4.7	30.4	21.6	ns	25.0	15.9	45.8	21.5	ns	34.0	13.8

Abbreviations: min: Minute; ns: Non-significant.

Table 9. Stress-induced changes of PNSi, SNSi, and BSTRi

	(A) Baseline		P	(B) Medium stress		P	(C) High stress		P
	Mean	SD		Mean	SD		Mean	SD	
PNSi	-1.4	0.8	#	-2.8	1.1	*	-3.8	0.7	
SNSi	1.9	1.5		7.6	6.4	°	12.5	5.6	§
BSTRi	14.0	5.9		30.4	21.6	°	45.8	21.5	&

Note: Average values of 16 police officers calculated from 2-min time segments are shown. # $P < 0.05$ A vs. B and vs. C; ‘&’ denotes $P < 0.05$ C vs. A; ‘§’ denotes $P < 0.05$ C vs. A and B; * $P < 0.05$ B vs. C; $P < 0.05$ B vs. A.

($R^2 = 0.98$), whereas the inverse relationship between PNSi and either SNSi or BSTRi was better modeled by NL functions (Figure 3B). The type of non-linearity between PNSi and SNSi appears to depend on whether the participant is experiencing physical or psychophysiological stress. The NL regression fitting PNSi and SNSi with a quadratic polynomial yielded an R^2 value slightly lower than that obtained from a cubic regression (from $R^2 = 0.978$ to $R^2 = 0.965$). In contrast, when analyzing physical stress data, the decrease in R^2 was more pronounced (from $R^2 = 0.961$ to $R^2 = 0.920$) (Figure 3A). Moreover, for SNSi

values between 10 and 20 units, PNSi values during effort and psychophysiological stress differed by nearly one unit on average. A similar trend was noted in the relationship between PNSi and BSTRi.

3.3.4. Relationship between Kubios indexes and HRV spectral parameters

The Pearson correlation between the PNS, SNS, and BSTR indexes calculated by Kubios and the HRV spectral parameters is presented in Table 10. Although significant ($P < 0.01$), the correlations between each index and the VLF, LF, HF, and Totalpower spectral components were moderate during daily activity and physical effort (R values ranging from 0.470 to 0.668) and moderate to strong during psychophysiological stress. Notably, there was no correlation with the LF/HF ratio in any situation.

4. Discussion

The dynamic nature of CAM is essential for maintaining physiological homeostasis, adapting to environmental demands, and ensuring proper responses to acute stress, thereby supporting survival in life-threatening

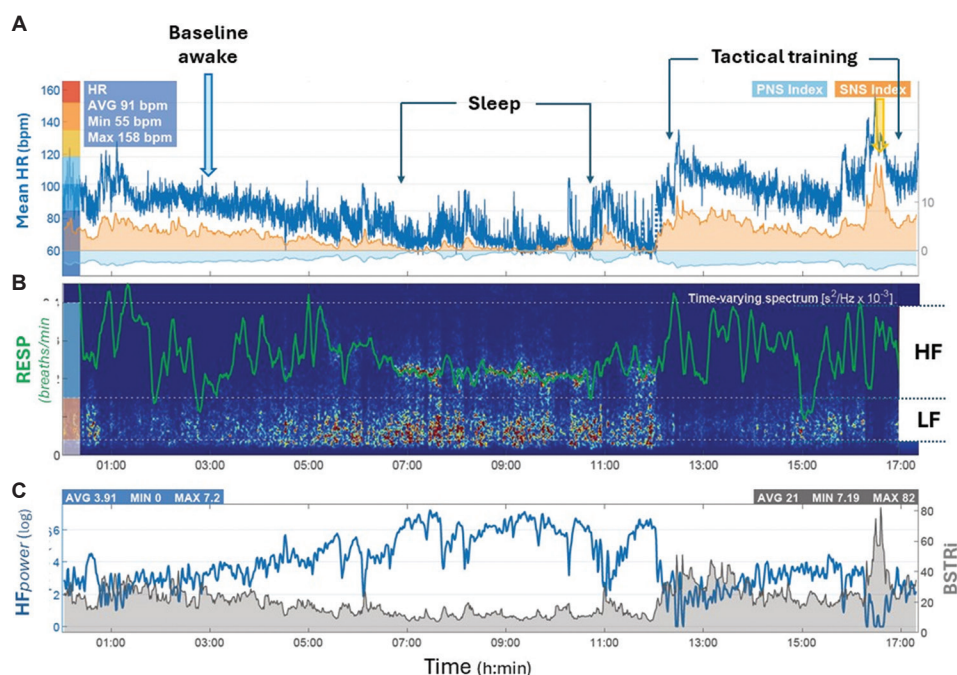


Figure 5. 24-h Holter recording from a police officer, including a realistic tactical training session. (A) Time-varying behavior of the mean heart rate, SNSi (orange area), and PNSi (light blue area). (B) Time-varying behavior of respiration (green line) and HRV spectral components (HF and LF). (C) Time-varying behavior of HFpower (blue line) and BSTRI (gray area), which temporarily reached a value of 82, accompanied by a drop in the HF spectral component

Abbreviations: PNS: Parasympathetic nervous system; SNS: Sympathetic nervous system; BSTRI: Baevski stress index; HR: heart rate; HF: High Frequency; LF: Low Frequency; RESP: Respiration.

situations.^{1,2,5,35} HRV analysis serves as a powerful non-invasive tool or assessing ANS function, deepening our understanding of CAM across various psychophysiological contexts. However, identifying which HRV parameters can serve as definitive markers of human stress responses and evaluating the suitability of different analytical methods has been widely debated.^{4,6-8,48} Notably, the use of LF/HF ratio as an indicator of “sympathovagal balance”^{6,7} has been called into question,⁸ with criticisms aimed at the rigid framework based on frequency bands.^{49,50}

In this study, a preliminary evaluation showed that short-term HRV analysis from 2-min intervals, which is useful for assessing transient CAM changes (such as those triggered by sudden acute stress), did not significantly alter the quantitative estimates of most HRV parameters (except for the recurrence plot) (Table 2). It also confirmed established differences between regular daily activity and NREM sleep (Table 3), as well as the age-related decline in HRV, regardless of whether linear or NL methods were used (Table 4).

Although there are known correlations between certain HRV parameters – such as TD SDNN with spectral Totalpower and VLF/LF components (sympathetic indexes) and TD RMSSD, pNN50 with spectral HFpower (parasympathetic

indexes) – the manual integration of multiple TD, FD, and NL HRV parameters remains challenging and time-consuming.⁴⁴ Many years ago, several indexes (e.g., the index of regulatory system activity) were proposed in space medicine to simplify the complex HRV-based evaluation of individual functional states.^{43,44} With the recent availability of automatically calculated indexes like PNSi, SNSi, and BSTRI,⁴⁰ this retrospective study aimed to evaluate their normality range in a healthy population, considering variability due to aging and situational factors such as normal daily activity, NREM sleep, physical effort, and real-world psychophysiological stress.

Notably, all three Kubios indexes incorporate non-HRV spectral parameters, making them potentially useful for independently assessing the functional significance of HRV spectral components in these contexts. Although a moderate but significant correlation was observed between each index and the VLF, LF, HF, and Totalpower, no correlation was found between the indexes and the LF/HF ratio (Table 10). This supports the ongoing criticism regarding the use of the LF/HF ratio as a definitive measure of sympathovagal balance.⁸

In recent decades, greater attention has been given to the FD VLF band due to its potential correlation with

Table 10. Pearson's correlation coefficients between PNSi, SNSi, and BSTRi and the HRV spectral components during baseline daily activity, physical effort, and psychophysiological stress

	PNSi	SNSi	BSTRi
Baseline			
VLFpow FFT ms^2	0.470**	-0.497**	-0.533**
LFpow FFT ms^2	0.492**	-0.530**	-0.626**
HFpow FFT ms^2	0.549**	-0.484**	-0.545**
TOIpow FFT ms^2	0.557**	-0.575**	-0.668**
LF/HF ratio FFT	-0.132	0.018	-0.033
Physical effort			
VLFpow FFT ms^2	0.510**	-0.465**	-0.468**
LFpow FFT ms^2	0.681**	-0.638**	-0.638**
HFpow FFT ms^2	0.579**	-0.659**	-0.660**
TOIpow FFT ms^2	0.599**	-0.591**	-0.602**
LF/HF ratio FFT	0.031	0.036	0.051
Psychophysiological			
VLFpow FFT ms^2	0.596**	-0.644**	-0.624**
LFpow FFT ms^2	0.787**	-0.756**	-0.727**
HFpow FFT ms^2	0.636**	-0.627**	-0.611**
TOIpow FFT ms^2	0.805**	-0.779**	-0.750**
LF/HF ratio FFT	-0.147	0.190	0.209

Note: ** $P < 0.01$.

Abbreviation: FFT: Fast Fourier transform.

stressful stimuli.^{19,20} In our study, VLFpower was higher during daily activity than during NREM sleep across all age groups, confirming its association with homeostatic mechanisms such as thermoregulation, baroreflex activity, and the renin-angiotensin system.²⁰ However, although significant, the correlations found between VLFpower and PNSi (positive correlation) and with SNS and BSTR indexes (negative correlation) were only moderate (Table 10).

Notably, a recent study reported a time-varying increase in VLFpower that followed the same trend as heart rate and the Kubios Baevsky stress index,⁵¹ suggesting that VLFpower may serve as a marker for stress-related increases in intrinsic cardiac sympathetic activity.^{21,52} However, the VLF spectral component is generally viewed as indicative of slower physiological mechanisms, sympathovagal balance, and a reduction in vagal tone during mental stress.^{20,21,53} The discrepancy between that study and our findings may be attributed to the VLF filtering effect of the detrending with the smoothness priors function used in our study.

Despite the notable changes in linear parameters associated with the well-known decline of HRV due to aging,¹² average PNSi values remained within the suggested normal range during both daily activity and NREM sleep.

Average SNSi values were slightly above normal during regular daily activity and, as expected, significantly lower during NREM (Table 5), with no significant age-related variations observed (Table 6). In contrast, the BSTRi was significantly higher in Age3 compared to both Age1 and Age2. This unexpected result may indicate that “healthy elderly” individuals exhibit a more complex adaptation of CAM, potentially influenced by a moderate increase in physiological stress. However, further research involving a larger population is needed to confirm this preliminary observation.

Although the evaluation of PNSi and SNSi was not affected by the duration of the time segments used for calculation, BSTRi values during high stress were higher (though not significantly) when calculated from 2-min intervals (Table 8), which may be more suitable for assessing the complex brain–heart interaction during critical events. However, we believe that the assessment of Kubios' BSTRi “normality range” requires further investigation. Notably, an interesting finding from this study, which has not been reported previously, is that average BSTRi values around 45 and even exceeding 80 conventional units were observed at peak stress during police tactical training. This suggests a variable combination of physical and psychophysiological stress, with individual peaks exceeding 60 in five cases and above 80 in one case (Figure 5). If squared (since Kubios' stress index is the square root of Baevsky's values), the observed values of 45–80 would correspond to 2025–6400 Baevsky conventional units, which are extremely high and theoretically inconsistent with a healthy status and adequate functional capabilities.^{43,44} This was not the case for our police officers; however, in a few instances, values exceeding 60 during highly stressful tactical scenarios were linked to poor tactical performance. Moreover, similar values were consistently achieved and tolerated by a 75-year-old volunteer during intensive effort without any subjective clinical symptoms or functional or ECG changes. Furthermore, as suggested by space medicine, individual physiological responses in critical situations may not align with average statistical normality, as adaptive reactions can vary based on individual psychophysiological and functional capabilities.⁴³ Interestingly, previous reports indicated that a similar BSTRi value in a highly fit police officer, without significant physical effort, was linked to a loss of situational control, resulting in operational failure and the sudden onset of paroxysmal arrhythmia, attributed to a marked acute CAM imbalance.⁵¹ Under such intense and uncontrolled psychophysiological stress, which can lead to immobilization (freezing) and an inability to respond to external threats, HRV decreases significantly. An unexpected increase in the delayed HF component has been noted in ultra-short-term HRV analysis,³⁵ aligning

with Porges' "polyvagal theory," which suggests that the *unmyelinated vagus* (the most primitive parasympathetic pathway), is activated when other defensive strategies (the *myelinated social vagus* and the *sympathetic* "fight or flight" response) fail to manage behavioral and physiological adaptation in life-threatening situations.^{23,54}

According to the "neurovisceral integration model," the interaction with the complex "brain–heart–body system," which allows for flexibility and adaptability to stressors, can be assessed through HRV analysis. Specifically, the HF spectral component serves as a reliable parasympathetic biomarker for measuring self-regulation and health.^{27,28,55}

A significant challenge in psychophysiology is quantifying the extent of stress-induced psychophysiological overload with HRV analysis⁵⁶ and its correlation with adaptive or maladaptive behaviors. The previously unreported finding that, under stress, the inverse correlation between the PNSi and the SNSi (and BSTRi) is NL (Figure 3), rather than the usual linear relationship at rest, could introduce a new framework for evaluating sympathovagal interaction in complex real-life critical situations. This may help define a critical level of dysfunctional parasympathetic modulation that impairs the "brain–heart–body system's" flexibility, resulting in insufficient rational control of stressful situations. This is supported by two additional findings indicating a significant mechanistic difference in CAM based on the type of stress encountered: (1) Although the relationship between the two indexes is NL in both cases, the nature of the non-linearity differs, with cubic functions providing a better fit for physical stress than quadratic functions. (2) For SNSi values between 10 and 20 units, the level of parasympathetic withdrawal was greater under psychophysiological stress, averaging nearly one SD of PNSi. If confirmed through targeted experimental studies, examining the dynamic relationship between PNSi and SNSi could serve as a valuable tool for identifying individual psychophysiological stress levels, which may influence the success or failure of operational outcomes, particularly in complex real-world situations that also involve physical effort, as seen in sports competitions or tactical operations.

Finally, the time-varying calculation of the PNS, SNS, and BSTR indexes effectively provided a quick overview of how CAM adapts to increasing physical effort (Figure 4) and the temporary rise in sympathetic drive alongside parasympathetic withdrawal during realistic tactical training (Figure 5). Further research is needed to determine if these additional parameters can enhance the ability to differentiate between psychophysiological stress and concurrent physical strain during real-world critical events.^{36,37}

5. Conclusion

The automatic calculation of PNSi, SNSi, and BSTRi indexes provides a quicker and more comprehensive assessment of adaptive CAM in both physiological and pathological conditions. Monitoring these indexes over time and in real time could greatly improve our understanding of heart–brain interaction dynamics, especially during acute stress from real-life critical events.⁵¹

In this study, PNSi and SNSi remained within the suggested normality range⁴⁰ at rest, with their variations during physical and psychophysiological stress showing the expected inverse correlation. If further research confirms that the inverse correlation between PNSi and SNSi (and BSTRi) under stress is NL and better fitted by NL functions (Figure 3), it could introduce a new approach for assessing sympathovagal interaction in complex real-world situations.

Given the relatively small sample size in this pilot study, although the results are promising, further research involving a larger population under dynamic physiological conditions is necessary to establish a reliable and potentially unbiased normality database. The BSTRi values calculated using Kubios software did not match those derived from the original Baevsky formula. Squaring the values obtained from this study of healthy individuals under effort or psychophysiological stress resulted in units that exceeded the range suggested by Baevsky, even for critical cardiac conditions.^{43,44} Therefore, the evaluation of the normality range for BSTRi calculated with Kubios software requires further investigation.

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Conflict of interest

The authors declare they have no competing interests.

Author contributions

Conceptualization: Donatella Brisinda, Riccardo Fenici

Investigation: Donatella Brisinda, Riccardo Fenici

Methodology: Donatella Brisinda, Riccardo Fenici, Marco Picerni

Formal analysis: Donatella Brisinda, Riccardo Fenici, Marco Picerni

Writing–original draft: Donatella Brisinda, Riccardo Fenici

Writing–review & editing: All authors

Ethics approval and consent to participate

Not applicable, being the study a retrospective analysis of ECG database data available from previous investigations carried out on a volunteered basis and/or for sports or duty fitness evaluation. All subjects had given verbal informed consent for the performance of their ECG recordings and analysis.

Consent for publication

All subjects had given verbal informed consent for the eventual use of their anonymized data for scientific and publication purposes.

Availability of data

The original clinical data are not publicly sharable due to ongoing privacy norms.

Further disclosure

A partial analysis of the 75-year-old volunteer training sessions was accepted as a poster presentation at the European Congress of Cardiology 2021.

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