

# ORIGINAL PAPER

International Journal of Occupational Medicine and Environmental Health 2024;37(1):84–97 https://doi.org/10.13075/ijomeh.1896.02272

# ANALYSIS OF THE RELATIONSHIP BETWEEN EMOTION INTENSITY AND ELECTROPHYSIOLOGY PARAMETERS DURING A VOICE EXAMINATION OF OPERA SINGERS

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#### Abstract

Objectives: Emotions and stress affect voice production. There are only a few reports in the literature on how changes in the autonomic nervous system affect voice production. The aim of this study was to examine emotions and measure stress reactions during a voice examination procedure, particularly changes in the muscles surrounding the larynx. Material and Methods: The study material included 50 healthy volunteers (26 voice workers - opera singers, 24 control subjects), all without vocal complaints. All subjects had good voice quality in a perceptual assessment. The research procedure consisted of 4 parts: an ear, nose, and throat (ENT)-phoniatric examination, surface electromyography, recording physiological indicators (heart rate and skin resistance) using a wearable wristband, and a psychological profile based on questionnaires. Results: The results of the study demonstrated that there was a relationship between positive and negative emotions and stress reactions related to the voice examination procedure, as well as to the tone of the vocal tract muscles. There were significant correlations between measures describing the intensity of experienced emotions and vocal tract muscle maximum amplitude of the cricothyroid (CT) and sternocleidomastoid (SCM) muscles during phonation and non-phonation tasks. Subjects experiencing eustress (favorable stress response) had increased amplitude of submandibular and CT at rest and phonation. Subjects with high levels of negative emotions, revealed positive correlations with SCM<sub>max</sub> during the glissando. The perception of positive and negative emotions caused different responses not only in the vocal tract but also in the vegetative system. Correlations were found between emotions and physiological parameters, most markedly in heart rate variability. A higher incidence of extreme emotions was observed in the professional group. Conclusions: The activity of the vocal tract muscles depends on the type and intensity of the emotions and stress reactions. The perception of positive and negative emotions causes different responses in the vegetative system and the vocal tract. Int J Occup Med Environ Health. 2024;37(1):84-97

#### Key words:

electromyography, job stress, autonomic nervous system, heart rate variability, singers, phonation

Funding: this study was realized within the project "Multimodal system for support of personalized therapy using metrorhythmic stimulation and walking with poles RAS 4 NoW" (grant No. POIR.01.01.01-00-0261/21).

Received: July 10, 2023. Accepted: December 12, 2023.

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# INTRODUCTION

An indispensable element of any emotional process is affect, which arises automatically and involuntarily as a result of arousal of the amygdala in 2 ways – through impulses originating directly from the thalamus (lower pathway) and through the cerebral cortex (upper pathway). These pathways reflect different emotional processes. One is a process of which the individual is not necessarily aware, while in the other there is secondary arousal of affects [1]. Physiolog-ically, the organism decides whether a situation is stressful or not; if it is, the autonomic nervous system (ANS) is then activated to initiate a stress response [2,3].

Stress itself is an anxiety-related experience [4]. Anxiety, as an emotional state or trait biologically is attributed to the amygdala, which is responsible for activating the eccrine sweat glands [5]. Activity of the glands on the palms of the hands and soles of the feet is associated with sweating, which is related to the emotions felt, as these areas are more reactive to psychological stimuli than to temperature. Sweat is a salty solution and gives rise to changes in skin conductivity and the generation of higher electrodermal activity (EDA) [6]. Electrodermal activity is a commonly used signal for behavioural-physiological analysis, reflecting both normative and pathological states [7]. Stimulation of the nervous system also affects cardiac function, which can be reflected in signals such as blood volume pulse (BVP) or heart rate (HR) [8], which again can be used to characterise emotional states [9]. In addition, heart rate variability (HRV) has been shown to serve as a good physiological indicator of the way the heart adapts to changing environmental conditions, both internal and external [10]. It seems reasonable to expect that HRV will be related to an individual's subjective emotional well-being [11].

These methods of monitoring emotion levels can contribute to a deeper understanding of the emotional and cognitive mechanisms [12–14]. This physiological data provides insights into psychological processes that would be difficult to discover using behavioural data alone [15]. In addition, the benefit of using physiological data is that it measures a phenomenon while it is occurring, rather than, as in the case of self-report methods, afterwards [16]. The standard way of measuring and analysing emotions is through self-report questionnaires. The fact that the data on experienced emotions then derives from the individuals themselves is, for many authors, an argument for the weakness of these methods. On the other hand, the individual is the expert on themselves, and no one else can gauge their feelings and assessments better. The description of the patient's emotional state is declarative, and although its evaluation is collected using validated psychological methods, it is subjective.

There is therefore a growing demand for complementing the traditional assessment of emotional states using objective analysis of physiological data. This creates an independent method to validate the patient's condition and maximise the accuracy of the diagnosis. Behaviours such as gestures and movements, together with physiological data (signals from the brain, heart, muscles, and skin), are non-verbal measures from the human body that very often reflect what people are thinking and feeling [17,18]. The combination of behaviours and physiological signals makes it possible to accurately describe human behaviour. In addition, computer-aided diagnosis and mathematical models make it possible to develop objective emotion recognition systems [19–21].

Reports in the literature indicate that the activity of the ANS may have a direct effect on the muscles of the vocal tract (the supraglottal and subglottal muscles) [22]. This relationship applies to both voice professionals and non-voice workers. But the interaction between ANS activity, emotional characteristics, and the voice – commonly regarded as a mirror of a person's internal state – is little understood [23]. The relationship between changes in ANS activity during vocal fold phonatory movements and what the person is actively thinking about during voice production raises questions about how different sorts of emotions affect ANS parameters [24,25]. As suggested by Benarroch [8] and



BVP – blood volume pulse; EDA – electrodermal activity; ENT – ear, nose, and throat; JAWS-1 – *Job-related Affective Well-being Scale* (JAWS) value after voice test; JAWS-2 – JAWS value after the hearing test; sEMG – superficial electromyography.

Figure 1. Flow diagram of the research protocol in the study on emotions and stress reactions during a voice examination procedure in 50 healthy subjects without vocal complaints, Institute of Physiology and Pathology of Hearing, Audiology and Phoniatrics Clinic, Poland, July–August 2021

Jerritta et al. [9], EDA in combination with HRV parameters and BVP, offers the possibility of a forming a multidimensional assessment of human emotional responses.

In this context, we should remember that the voice examination procedure itself may evoke different emotions in a subject [26]. It can be a source of social anxiety associated with assessing oneself, and in voice professionals it may induce stress from the feeling that their professional competence is being judged [22,27]. Bugdol et al. [28] recently advocated that the procedure of evaluating voice professionals should be modified, and any ear, nose and throat (ENT) examination should be supplemented with questionnaires and standardized methods for assessing emotions and stress.

#### Purpose

This study aimed to analyse emotions and stress reactions during a voice examination procedure and assess correlations with activity of the laryngeal muscles involved in speech.

# MATERIAL AND METHODS

The study material included 50 healthy subjects without vocal complaints. The study group consisted of 26 professional opera singers (11 men, 15 women). The mean age in this group was 40 years (SD = 10). The control group included 24 healthy volunteers (12 men, 12 women). All subjects had good voice quality in a perceptual assessment, no his-

tory of concurrent laryngeal or systemic disease, and normal hearing. The mean age in this group was 43 years (SD = 12). After being informed about the nature of the investigation, each person gave voluntary written consent to participate in the study. All participants were provided written informed consent before the beginning of testing. The Bioethics Committee of the Institute of Physiology and Pathology of Hearing in Warsaw approved the study protocol (No. KB.IPFS 1/2021) and conducted it according to the Declaration of Helsinki. Authors confirm that all methods were performed in accordance with the relevant guidelines and regulations.

The research procedure consisted of 4 parts: an ENT-phoniatric examination, superficial electromyography (sEMG), recording of physiological indicators (HR and skin resistance) using a wearable wristband (Empatica E4, Boston, USA), and psychological profile based on the answers to the questionnaires (1-item to assess the anticipated stress, and the intensity of experienced emotions *Job-related Affective Well-being Scale* (JAWS), used twice during the research after the voice test [JAWS-1], and the hearing test [JAWS-2]). The entire research protocol is shown in Figure 1.

#### Physiological measurement

Endoscopic examination of the larynx was performed through the nose, using a Xion fiberscope with a diameter of 3.2 mm and a rigid endoscope.

After the physical examination, each subject was fitted with a certified Empatica E4 device allowing real-time recording of physiological signals encompassing HRV, EDA, temperature, BVP, and accelerometric signal (ACC). The continuous wristband signal was recorded throughout the test procedure (Figure 1), and then analysed in whole and in terms of segments during the voice and hearing test. Certain indices were calculated at a later stage using MATLAB software. Basic parameters and their statistics were calculated for each signal according to standard methods [29]. For the EDA signal, the following were determined: mean (M), minimum (min.), maximum (max), standard deviation (SD), 25th and 75th percentile, skin conductance, tonicity, additive error, number of galvanic skin responses (GSRs), GSR amplitude, and GSRs per minute (GSR/min). From BVP we calculated bandpass, centre of gravity (CoG), and frequency. The HRV parameters determined were: SD of intervals between successive heartbeats (R-R intervals) differences (SDSD), SD of NN intervals (SDNN), root mean square of successive RR interval differences (RMSSD), percentage of successive RR intervals that differed by >50 ms (pNN50), integral of the density of the RR interval histogram divided by its height (triangular index - TRI), baseline width of the RR interval histogram triangular interpolation of the NN interval (TINN), and HR.

The sEMG examination included the simultaneous recording of neck potentials using a 4-channel EMG device (Neurosoft, Ivanovo, Russia). Measures were made of:

- submentalis muscles (SUB) (the anterior belly of digastricus and geniohyoid),
- left cricothyroid muscle (CT) (together with the overlying superior belly of the sternohyoid muscle),
- left and right sternocleidomastoid (SCM) muscles,
- SCM symmetry (difference between right and left SCM amplitudes) [30].

The recording procedure followed the diagnostic standard published in the literature [31,32]. The authors evaluated

sEMG mean and max amplitude during different phonatory and non-phonatory tasks – at rest, saliva swallowing, free phonation [a], and glissando [a] (from low to high frequencies).

## Psychological assessment

Psychological measures were made using a paper-andpencil test. Anticipatory stress was assessed before the study with a single question, "Please indicate to what extent, from 1 to 10, do you feel stressed about testing your professional competence during the ongoing study?", which was modelled on a visual analogue scale (VAS). Emotional intensity was assessed using the standardised JAWS [33]. Since JAWS was originally designed to examine emotions experienced in the working environment the instruction of scale was adapted for the research protocol. The items concerned the intensity of particular emotions during activity of research participant in the voice and hearing tests, respectively. The version used in the study consists of 12 items. A 5-point Likert scale was used (1- never, 5 - very often). The reliability of the tool used in the study was calculated using the Cronbach's a. For the overall result for JAWS-1 its value was 0.735 (min.-max 0.639-0.810), for the subscale of positive emotions 0.677 (min.-max 0.549-0.775), and for negative emotions 0.752 (min.-max 0.659-0.824). For the overall result for JAWS-2 its value was 0.752 (min.max 0.659–0.824), for the subscale of positive emotions 0.702 (min.-max 0.580–0.794), and for negative emotions 0.761 (min.-max 0.661-0.836). These results can be considered satisfactory as they exceeded the recommended value of 0.70. Each person completed the questionnaire twice, after the voice test (JAWS-1) and after the hearing test (JAWS-2), marking the answers immediately after the test. Based on the results in the questionnaires, 4 values were calculated for each person, corresponding to the different categories of emotional states experienced. The categories, according to the theory of van Katwyk,

take into account differences in the intensity of arousal (low-high level of activation) and the sign of the emotion (pleasant [positive] – unpleasant [negative]) [33]. The categories formed the following combinations: pleasant and intense (high pleasure – high arousal, e.g., joy), unpleasant and intense (low pleasure – high arousal, e.g., anger), pleasant with low intensity (high pleasure – low arousal, e.g., satisfaction), and unpleasant with low intensity (low pleasure – low arousal, e.g., boredom). Integrating van Katwyk's views with the classical stress theory, according to Selye, positive emotions with high activation can be called eustress (psychological eustress). In contrast, negative emotions with a high degree of intensity can be called distress (psychological distress) [33,34].

Eustress occurs in response to stressful stimuli and is a reaction that is conducive and beneficial to an individual's health. A feature of this response is the feeling of emotions, i.e., excitement, joy, increased energy for action, and improved performance. In contrast, distress is associated with unpleasant emotions, i.e., anxiety and frustration, which leads to cognitive distortions (deficits in attention, memory, reasoning), errors and mistakes in performance, and sometimes leads to withdrawal from action. The obtained eustress–distress values describing experienced emotions were correlated with the values of vocal tract muscle amplitudes during various phonation and non-phonation tasks.

#### Statistical analysis

The results of the study were subjected to statistical analysis. The analyses investigated the dependence of resting and action potential values of the examined muscles surrounding the larynx on the physiological parameters during the completion of individual questionnaires, the psychological profile relating to reactions presented during task performance – mobilizing stress (eustress) and paralyzing stress (distress). Differences between the group of voice professionals and the controls were then examined. On this basis, correlations between muscle activity of the vocal tract and subjective assessment of the subjects were obtained.

The next stage of the analysis concerned relating the results to objective values of ANS activity parameters. Statistical analysis involved comparing samples using a Mann-Whitney U test because, in most cases, the normality (Shapiro-Wilk test) or the variance equality assumption (Bartlett's test) were violated, and this choice enabled inference consistency. If statistical significance was achieved, a Glass rank biserial correlation coefficient, indicating the effect size, was calculated. This paper does not report small effects since they are not clinically relevant. In all statistical calculations, the significance level was set to 0.05.

## RESULTS

Statistical analysis revealed significant correlations between the intensity of experienced emotions and vocal tract muscle amplitudes, during both phonation and non-phonation tasks. A correlation was obtained between stress and the difference between SCM<sub>max</sub> at rest (p < 0.05, r = 0.38) and during phonation (p < 0.05, r = 0.32). Similarly, there was a correlation between stress and the difference between  $CT_{max}$  at rest during free phonation (p < 0.05, r = 0.31) and during glissando (p < 0.05, r = 0.35). Details of the correlation coefficients are given in Tables 1.

It was observed that the activation of extremely different emotions (positive and negative) and their intensity coexisted with different vocal tract muscle tone strength. The findings are illustrated in Table 2.

Subjects experiencing eustress (positive JAWS), i.e., a favourable stress response, had increased amplitude of SUB and CT at rest. This relationship was observed in both the JAWS-1 and JAWS-2 results (collected after the voice and hearing tests, respectively), which may indicate a active and positive attitude towards the assessment of

Emotion				Correlation				
	l-uou	phonation task				phonation	task	
	rest		saliva swallc	owing	free phonati	u	glissando	
	~	d	L	d	7	d	L	d
JAWS-1								
total		>0.05		>0.05		>0.05	SCM <sub>difference max</sub> —0.31	
positive	SUB mean 0.35, CT mean 0.32, CT max 0.32		SUB <sub>mean</sub> -0.37			>0.05		>0.05
high positive	SUB <sub>mean</sub> 0.38, SUB <sub>max</sub> 0.33, CT <sub>mean</sub> 0.38, CT <sub>max</sub> 0.38		SUB <sub>mean</sub> -0.42		SUB <sub>mean</sub> 0.36, SUB <sub>max</sub> 0.38, CT <sub>max</sub> 0.36, SCM <sub>maxleft</sub> 0.34, SCM <sub>maxright</sub> 0.36, SCM <sub>difference max</sub> 0.31			>0.05
low positive		>0.05		>0.05		>0.05	SCM <sub>difference max</sub> —0.31	
negative		>0.05		>0.05		>0.05	SCM difference max 0.36	
high negative		>0.05		>0.05		>0.05	SCM difference max 0.56	<0.01
low negative		>0.05		>0.05		>0.05		>0.05
JAWS-2								
total		>0.05		>0.05		>0.05	CT <sub>max</sub> 0.32, SCM <sub>difference max</sub> -0.36	
positive	SUB mean $0.37$ , CT mean $0.34$ , CT mean $0.34$ , CT mean $0.35$			>0.05		>0.05	CT <sub>max</sub> 0.34	
high positive	CT <sub>mean</sub> 0.34, CT <sub>max</sub> 0.34			>0.05		>0.05		>0.05
low positive	SCM <sub>difference mean</sub> —0.39 SCM difference mean —0.39			>0.05	CT <sub>mean</sub> 0.35		CT <sub>max</sub> 0.41	
negative		>0.05		>0.05		>0.05	SCM difference max 0.34	
high negative		>0.05		>0.05		>0.05		>0.05
low negative		>0.05		>0.05		>0.05	SCM difference max 0.33	

**Table 1.** Statistically significant correlations (p < 0.05) between muscles tested during different phonation and non-phonation tasks and *Job-related Affective Well-being Scale* (JAWS) value

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Emotion		Muscle activation			
EIIIOUOII	rest	phonation	swallowing		
Eustress	↑ SUB, CT	↑ SUB, CT	$\downarrow$ SUB		
Distress	_	$\uparrow$ SCM <sub>max</sub> (glissando)	$\uparrow$ SCM <sub>max</sub> (glissando)		

**Table 2.** Schematic representation of the relationship between activation of muscle groups surrounding the larynx

 and different emotions (eustress and distress) in 50 healthy subjects without vocal complaints, Institute of Physiology and Pathology of Hearing,

 Audiology and Phoniatrics Clinic, Poland, July–August 2021

 $\uparrow$  increase;  $\downarrow$  decrease.

Abbreviations as in Table 1.

professional competence. This group of subjects was also characterized by lower SUB tension during reflex actions (swallowing saliva).

Comparing the scores of JAWS-1 and JAWS-2 for the intensity of highly activated positive emotions (high positive), it was noted that the JAWS-1 score correlated with increased muscle amplitude, not only at rest but also during phonation. Subjects scoring high in this category activated the SCM muscles more strongly. Subjects in whom the situation evoked intense positive emotions had positive correlations with the amplitudes of SUB and CT at rest and phonation and negative correlations of SUB with swallowing. This indicates higher muscle activity at rest and during phonation tasks, and greater muscle relaxation during reflex actions (swallowing).

Subjects with high levels of activation about negative emotions (low pleasure – high arousal), i.e., those who experienced paralyzing stress in response to the assessment situation, revealed positive correlations with SCM<sub>max</sub> during the glissando, both in the assessment of emotions after the voice and hearing test (JAWS-1 and JAWS-2). In addition, subjects had a stronger correlation with JAWS-1. The tendency to distress during task performance co-occurred with SCM activation. Sternocleidomastoid activation indicates stimulation of additional respiratory muscles, which should not be activated under conditions of average voice production. Individuals activating positive emotions in stressful situations (high-pleasure) showed significantly less asymmetry of SCM with increased vocal effort (glissando) than negative (low-pleasure) individuals. The relationship was observed in both the JAWS-1 and JAWS-2 questionnaire scores.

The above results were analyzed in terms of differences in the group of singers compared to the control group. A higher incidence of extreme emotions was observed in the professional group. Significant differences were only obtained for "low pleasure – high arousal" emotions in the JAWS-1 questionnaire conducted immediately after the voice test. Singers had a higher mean score (M±SD 4.22±2.06) than the control group (M±SD 3.46±0.84). An almost- significant difference (p = 0.054) was obtained for the "high positive" value in the JAWS-1 test completed by singers. The professionals had a higher mean (M±SD 9.26±2.03) than the control group (M±SD 8.04±2.63). The results in both groups for high positive and high negative emotions are shown in Figures 2a and 2b.

The above results show the relationship between emotions related to voice testing with the activity of individual vocal tract muscles. In a subsequent analysis, the authors tried to relate the results to the objective values of ANS activity parameters recorded using the Empatica E4 device. Indeed, correlations emerged between the values of the physiological parameters and emotions engendered by the test. Table 3 shows statistically significant correlations between the values of sEMG amplitudes and individual electrophysiological parameters measured with



**Figure 2.** Comparison of *Job-related Affective Well-being Scale* (JAWS) value after voice test (JAWS-1) scores between singers and non-singers: a) distress – a statistically significant difference, b) eustress – the difference approaches significance (p = 0.054), in the study on emotions and stress reactions during a voice examination procedure in 50 healthy subjects without vocal complaints, Institute of Physiology and Pathology of Hearing, Audiology and Phoniatrics Clinic, Poland, July–August 2021

the Empatica 4 during the entire recording and during individual time segments.

Notably, the most significant correlations were recorded when subjects had just completed the questionnaire related to feelings about the voice (JAWS-1). The following correlations with HRV parameters were obtained during voice recording. Weak positive correlations were obtained between HRV parameters (SDSD, SDNN, RMSSD) measured during acoustic voice testing and the value of the amplitude (mean and maximum) of the CT muscle tone measured during glissando. In addition, during glissando, weak positive correlations were obtained between HRV parameters (pNN50, TRI, HR) and the maximum amplitude from the SUB muscles. At rest, weak positive correlations were obtained between HRV parameters (pNN50, TRI, HR) and the value of the difference in mean amplitude from the SCM muscles and the value of the difference in maximum amplitudes for the HRV parameter SDNN.

Correlations with EDA, BVP, and HRV parameters were obtained when subjects completed the JAWS-1 question-naire.

There were weak, moderate, and strong positive correlations between the EDA parameters recorded during the JAWS-1 questionnaire and the amplitudes of the muscles at rest, during swallowing, and during phonation. Mean positive correlations were obtained between EDA parameters and the amplitudes (mean and maximum) of the SUB muscles measured during rest, swallowing, and phonation (free and glissando). In addition, a strong correlation was registered between the min. EDA parameter and the difference in maximum amplitude from the SCM muscles at rest and during free phonation. A moderate correlation was registered with the mean and maximum values of SUB and CT amplitudes at rest and during phonation for the entropy EDA parameter.

Moderate positive correlations were obtained between the CoG BVP parameter recorded during the JAWS-1 questionnaire and the values of the mean amplitudes of the muscles SUB and CT measured during glissando. Weak to moderate positive correlations were obtained between the HRV parameters (SDSD, SDNN, RMSSD, pNN50, TRI, power spectral component in low frequency [pLF]) measured during the completion of JAWS-1 and the amplitude (M and max) of the CT muscles measured during free phonation. In addition, for the HRV parameter TRI, weak positive correlations were obtained between

A	Correlation				
Activity	rest	saliva swallowing	free phonation	glissando	
HRV					
voice examination	$SCM_{mean},SCM_{max}$	-	-	SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	
EDA				incur incu	
JAWS-1	$SUB_{mean}, SUB_{max}$	SUB <sub>mean</sub>	$SUB_{mean},SUB_{max}$	SUB <sub>max</sub>	
JAWS-2	$SUB_{mean}$ , $SUB_{max}$ $CT_{mean}$ , $CT_{max}$	-	$SUB_{mean}, SUB_{max}$ $CT_{mean/max}$	SUB <sub>max</sub>	
CoG BVP					
JAWS-1	-	-	-	SUB <sub>mean</sub> CT <sub>mean</sub>	
HRV				nican	
JAWS-1	-	-	SUB <sub>max</sub> CT <sub>max</sub> , CT <sub>max</sub>	-	
TRI			ilicali iliax		
JAWS-1	SUB <sub>mean</sub> , SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	-	$SUB_{mean}$ , $SUB_{max}$ $CT_{mean}$ , $CT_{max}$	-	
HR					
JAWS-1	-	SCM <sub>difference mean</sub>	-	SCM <sub>difference mean</sub>	
hearing test	-	SCM <sub>difference mean</sub>	SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	
JAWS-2	$SUB_{mean}$ , $SUB_{max}$ $CT_{mean}$ , $CT_{max}$	-	$SUB_{mean}$ , $SUB_{max}$ $CT_{mean}$ , $CT_{max}$	CT <sub>max</sub>	
entire registered signal	SUB <sub>max</sub> , CT <sub>max</sub>	-	SUB <sub>mean</sub> , SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	SUB <sub>max</sub> CT <sub>mean</sub> , CT <sub>max</sub>	

**Table 3.** Statistically significant (p < 0.05) correlations between physiological signals recorded with the Empatica E4 device and superficial electromyography (sEMG) amplitude of individual muscles in 50 healthy subjects without vocal complaints, Institute of Physiology and Pathology of Hearing, Audiology and Phoniatrics Clinic, Poland, July–August 2021

CT - cricothyroid; EDA - electrodermal activity; HRV - heart rate variability; SCM - sternocleidomastoideus; SUB - submentalis; TRI - triangular index.

JAWS-1 – Job-related Affective Well-being Scale (JAWS) value after voice test; JAWS-2 – JAWS value after the hearing test.

The simplified table sets out the type of parameter from which the correlation was obtained; the time segments of the recording during which significant correlations were obtained; and the type of muscles and type of activity.

HRV (pNN50, TRI, HR) and the maximum amplitude from the SUB muscles. Weak positive correlations were obtained at rest and during free phonation for the amplitude (M and max) from the CT muscles and the maximum amplitude from the SUB muscles. In addition, at rest and during glissandi, moderate positive correlations were obtained between the HRV HR parameter and the difference in mean muscle amplitude SCM. During the hearing test, the following correlations with HRV parameters were obtained. Weak to moderate positive correlations were obtained between HRV parameters (SDSD, SDNN, RMSSD, pNN50) measured during the hearing test and the value of the difference of the mean amplitude of the SCM muscles during saliva swallowing, as well as the mean and maximum value of the SUB muscle amplitudes and the maximum amplitudes of the CT measured during free phonation and glissando.

When subjects completed the JAWS-2 questionnaire, the following correlations with HRV parameters were obtained. Weak and moderate positive correlations were obtained between HRV parameters (SDSD, SDNN, RMSSD, pNN50, TRI, pLF, power spectral component in high frequency [pHF], LF/HF ratio) measured during the completion of JAWS-2, and the value of the amplitudes (mean and maximum) of the CUB and CT muscles measured during rest and free phonation, and for the values of the maximum CT amplitudes during glissando.

Correlations with HRV parameters were obtained throughout the recorded signal. Weak and moderate positive correlations were also obtained between HRV parameters (SDSD, SDNN, RMSSD, pNN50, TRI, HR) measured throughout the recording and values of maximum SUB and CT muscle amplitudes at rest, free phonation, and glissando, and values of mean SUB and CT muscle amplitudes during free phonation, as well as values of mean CT muscle amplitudes during glissando.

## DISCUSSION

Although stress has been frequently attributed to voice disorder development and progression, little work has been done to determine the role of activation of the ANS on voice production parameters [35]. In this study we focused on the analysis of emotions and stress reactions during a voice examination procedure and assess correlations with activity of the laryngeal muscles involved in speech. This work is of unique value, setting out the extent of different emotions accompanying voice testing and measuring the associated vocal tract muscle activity during phonation and non-phonation. The paper provides correlations of peri-laryngeal muscle activity with emotion in terms of the HRV, BVP, and EDA indices of ANS activity. So far, there have been no reports in the literature addressing this issue. The results of ENT-phoniatric examination of the study group including the analysis of medical history, endoscopic results, voice selfassessment, voice acoustic analysis and analysis the results of EMG muscle activity were published previously by the authors [30].

The methods used in this study were based on the diagnostic standard of the otorhinolaryngological and phoniatric examination. In order to achieve the objectives, the study was supplemented by an original algorithm for electromyographic assessment of the muscles surrounding the larynx, the recording of parameters of the autonomic system and the determination of psychological factors of the subjects. The limitation of the study is the small sample size of 50 healthy subjects without vocal complaints (26 professional opera singers and 24 healthy volunteers). The literature review for the research project showed that there have been papers describing changes in ANS parameters under stress [36]. Stress is also mentioned as one of the causal factors in voice disorders [37]. Thus, a multidisciplinary approach to voice professionals, especially opera singers, and measurement of multifaceted voice parameters, is essential. As stated by Cardoso, knowing the relationship between ANS, the laryngeal muscles, and the voice is important in developing a transdisciplinary approach to overcoming voice problems [38]. This study has shown that the emotions associated with voice testing correlate with altered tension of the peri-laryngeal muscles and variations in ANS parameters (EDA, BVP, and HRV). Increases in EDA (recorded after completion of the JAWS-1 questionnaire and during the subject's rating of their emotions in reaction to the voice test) were significantly more frequent in patients with increased tension in all studied conditions (resting, phonatory tasks, and non-phonatory tasks). Correlations of BVP parameters with JAWS-1 results and sEMG testing were most apparent during the intensification of vocal effort (glissando). The highest correlations were obtained for HRV parameters. The profile of increased vocal tract muscle activity was different for these parameters, occurring only in phonation and rest-related activities. Emotional arousal

from the voice test was stronger than during the hearing test and JAWS-2. These differences confirm the correlation of HRV with stress – HRV is the most sensitive parameter but it also follows a different course than the voice examination procedure, especially in voice professionals.

The analyses suggest that a more health-promoting stress response is increased SUB and CT muscle activity at rest and phonation. This was evident with each task performed during both auditory and voice testing. Depending on the type of stress response, the differential activation of the muscles surrounding the larynx is confirmed by observing different SCM activation. The paralyzing stress response was evident in SCM muscle activation with stronger phonation intensity. We found correlations with distress in studies related to voice assessment. The activation of the SCM, or auxiliary respiratory muscle during glissando, can be explained by emotionally driven accelerated breathing. According to Pettersen and Watson, activation of this muscle during phonation can only occur physiologically when there is extreme vocal effort during singing, such as with opera singers performing arias in the upper range of their voice [39,40]. As Van Puyvelde et al. points out, changes in activation of the muscle system associated with respiratory function under the influence of emotion, ANS activity, and stress might be the missing link in understanding the mechanisms between speech and stress [41].

In addition, individuals with a tendency towards intense emotional activation show greater muscle activity and tension. In individuals predisposed to negative emotions, stress leads to activation during phonation of inappropriate muscle parts surrounding the larynx. In contrast, this study did not assess ways of coping with stress. Based on the literature, an improved stress response might come from better ways of dealing with stress, such as through greater self-confidence and less stage fright [42].

There are papers in the literature showing different characteristics of vocal tract activity associated with voice training in professionals at rest and during phonation [4,22]. The present study also found that singers reported significantly more extreme emotions (both positive and negative) associated with the voice-training procedure. The results suggest that the voice assessment as a test of competence for singers was stressful and evoked more intense emotions than in non-voice workers. This can be seen in those who generated negative emotions (fear, anxiety) or in those who found it challenging. Activation of emotions may indicate the validity of the adopted research procedure, which was designed so that the tasks would trigger a behavioural response in the subjects which could then be assessed.

The voice is the most important means of human communication. Its role in modern civilisation has been on the increase in recent years and so has the number of professions dependent on the voice. One of the directions in the development of modern otolaryngology and phoniatry is the search for methods to objectify glottal function. The use of neurophysiological indicators to assess voice production is particularly important in the context of studying the effects of stress and emotions. There are reports in the literature of voice-dependent occupations that are associated with high levels of stress, such as teaching or working in call centres. It would be interesting to continue the research with larger samples, including other groups of professional voice users.

#### CONCLUSIONS

- The perception of highly different emotions (positive and negative) causes different responses of the vegetative system, altering muscle tension in the vocal tract.
- The voice testing procedure for voice workers is stressful, triggering specific emotions that affect the state of tension of certain vocal tract muscles.
- The activity of the vocal tract muscles depends on the type and intensity of the emotions and stress reactions.
- Standard methods for assessing emotions and stress should be used when examining voice workers.

#### Author contributions

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