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Real Time Mental Stress Detection through Breath Analysis

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Abstract. Modern work environment is changing from classic physiological to more psychological workloads. The capability to monitor biochemical processes within the human body positions breath analysis as a promising method to non-invasively and quickly detect mental stress. The present work aims to identify psychological stress biomarkers and breath profile changes after psychological interventions (PASAT or relaxing videos). In an exploratory study, 14 male participants followed a double cross-over randomized study which included two experimental sessions (stress and neutral/relax). GSR and HR were continuously measured to indicate physiological stress levels. NASA-TLX questionnaires were fulfilled to quantify individual stress. Breath samples were selectively collected by a recently developed advanced breath sampling prototype device and afterwards analyzed with a GC-IMS apparatus. Some promising results on mental stress detection were found and are presented and discussed in this paper

Keywords: Mental Stress, Volatile Organic Compounds (VOCs), Breath Analysis, Gas Chromatographic-Ion Mobility Spectrometer (GC-IMS) Paced Auditory Serial Addition Test (PASAT), Galvanic Skin Response (GSR), Heart Rate (HR), NASA Task Load Index (NASA-TLX)

1 Introduction

In the psychobiology field, stress is an unpleasant emotional state and the physic and/or psychologic result to a threatening situation. The frightening experience is caused by an imbalance between environmental demand for performance and available personal resources, which triggers behavioral, emotional and cognitive reactions [1]. In response to an acute threat to survival, a stress pattern is prompted (*e.g.* as “fight-or-flight response”) which is related to the release of stress hormones and, consequently, to the activation of specific muscles and other bodily functions. [2] Such process includes variations in cardiac activity, sweat gland activity, and skin temperature. A high number of physiological signals have been broadly used as reliable indicators of stress (*e.g.*

GSR, HR, skin temperature, electrocardiogram, electromyography, blood volume pulse, electroencephalogram and respiration rate) [3].

Multiple wearable biosensors combine and process such type of physiological data with high temporal resolution [4] and can be used for basic research, clinical application, or during daily routines in real-life situations [5]. However, their use in real-world experiments poses several challenges in terms of reliable and useful measurements for emotion extraction [5]. Those challenges include: selection of a suitable sampling frequency (to depict the signal correctly), proper sensor placement (for signal ambiguities avoidance) and the data treatment regarding the small fluctuations of the raw signals, caused by the oscillations of the physiological status and inadaptation of sensors to daily human behavior through measurements.

Therefore, there is still a need for innovative approaches that can detect stress in real-world settings while overcoming the aforementioned challenges.

Exhaled air is a wealthy and complex matrix with hundreds of compounds. Recent health diagnosis and monitoring research points breath analysis and “breathomics” as one of the highest potential research fields for systemic metabolic processes assessment. Thus, a research need arose to investigate whether stress may be identified by breath analysis and whether any such putative VOC markers might have the potential to both detect and differentiate stress levels.

This study was sought to generate a stress response in human breath using PASAT interventions [6]. As proposition, breath composition is nearly constant and variations only occur with perturbations (*e.g.* disease, behavioral and environmental), which changes metabolism and volatalome. The underlying hypothesis was that induced stress cause a faster breathing rate, an increased pulse rate and blood pressure which would be related with changes in exhaled VOCs across the participant cohort.

2 Contribution to Life Improvement

Mental health disorders are among the most burdensome health concerns in developed countries. Nearly 792 million people (10.7%) worldwide reported any mental illness type in 2017. Additionally, 71% of adults reported at least one stress symptom, such as headaches, depression or anxiety. Human and financial costs of stress are enormous: around 120,000 people die each year from work-related stress, 5 to 8% of healthcare expenses only in the US are due to work-related stress (over \$100 billion/year) [7].

In addition to the long-term effects of stress (*e.g.* increased stroke risk and negative influences on the cardiovascular system), work-related stress is also known to have a negative impact on work performance and concentration leading to burning out, confrontations and loss of productivity (with higher human error rates). Approximately 80% of aviation accidents are caused by pilot errors, a result of insufficient management of stress [8]. Therefore, the constant use of innovative and quick stress monitoring approaches in jobs with high responsibility and high stress levels (*e.g.* pilots or astronauts) remains crucial since it may prevent fatal accidents.

Breath profiling is innately attractive allowing noninvasive, quick and straightforward observations of multiple biochemical processes occurring in the human

body. Recently the non-invasive detection of biomarkers in breath associated with tuberculosis (TB), chronic obstructive pulmonary disease (COPD), asthma and an array of cancers have been reported [9] [10] [11]. A link between psyche, emotions and the composition of exhaled breath has also been found in recent studies [12]. However, the observation and description of the effect of psychological stress on the VOC profile from breath is currently almost unreported.

Although breath analysis presents many advantages, the lack of standardized methods to both collect and analyze breath samples are still unresolved issues. The present paper reports the results of the application of a protocol for mental stress detection [13], combining: (a) a recently developed selective and adaptative system for breath sampling [14] and, (b) a fast, reagent-free and portable analyzer (GC-IMS) with high sensitivity and selectivity (ppb_v - ppt_v range) [15].

3 Study Design & Methodology Applied

The study was conducted in accordance with the ethical principles of good clinical practice of the Declaration of Helsinki. Fifteen healthy male non-smokers volunteers (excluding any lung diseases, heart conditions, schizophrenia), aged 20-35 years, gave written informed consent to participate in the study. Other pre-conditions were also required: (a) 2-5 hours of fasting; (b) no consumption of caffeine or taurine containing products; (c) 2-5 hours from the last mouth wash; (d) minimum of 5 hours of sleep in the night before the measurements; (e) no use of hygiene products or perfume prior to the measurements and; (f) no heavy consumption of alcohol 24 hours before sessions. The presented study applied a previously described protocol for mental stress detection through breath analysis [13]. Such protocol includes the methodology specifications for stress inducement, the description of the equipment used for HR, GSR and breath acquisition and the data analysis performed [13]. Therefore, a randomized cross-over design study was implemented including two sessions (RS - neutral/relax and SS - stress), each comprising two stimuli (less and more intense, respectively).

Within SS, a standardized cognitive test (PASAT) was chosen to elicit stress [16], whereas RS included a relaxing video. Baseline measurements of breath (BS), heart rate (HR) and galvanic skin response (GSR) were collected before stimuli. Mid and long-term effects of stimuli were also assessed by measuring 5 minutes and 1 hour after the last exposure, respectively. NASA TLX were also fulfilled in the end of each campaign to evaluate subjective workloads felt within sessions.

Fourteen complete sets of 5 end-tidal breath samples (with a 5.0 cm³ volume) were collected: baseline (BS01), immediately after the first (BS02) and second stimuli (BS03), 5 min after BS03 (BS04) and 1 hour after (BS05). All samples were obtained using an advanced prototype for selectively breath sampling (depending on the origin, *i.e.* oral cavity, esophageal or alveolar) developed by NMT, S.A [14].

4 Results & Discussion

4.1 Cohort Homogeneity

The homogeneity of the study population was evaluated according to the age, body-mass index (BMI), sleeping and fasting time of participants of both groups (A and B). Both age and BMI do not present any significant change ($p\text{-value} > 0.05$) between groups (26.6 ± 5.1 years and 24.4 ± 4.2 , respectively). The fasting time of the participants from group A (3.6 ± 0.8 hours) has only proven to be significantly different ($p\text{-value} = 0.017$) from group B (2.6 ± 0.2 hours) within the neutral/relax session. Fasting time differences within the neutral session were not observed between sessions ($p\text{-value} = 0.569$). Sleeping time values present no significant differences on participants' cohorts within and between sessions. Since no significant or relevant differences between participants' cohorts were exhibited, onwards the population will be considered as a single group.

4.2 Heart Rate and Galvanic Skin Response

In order to verify the stress and relax condition of volunteers during the experimental procedure, traditional markers were used (HR and GSR). In Figure 1, individual HR and GSR temporal responses through stress (SS) and the neutral/relax sessions (RS) are shown. As expected, for the RS, HR almost does not change from the baseline threshold. During both stimuli, HR and GSR values are undoubtedly different between sessions, with higher response during stress application. HR and GSR variation were higher for stimuli 1 and 2 of the SS.

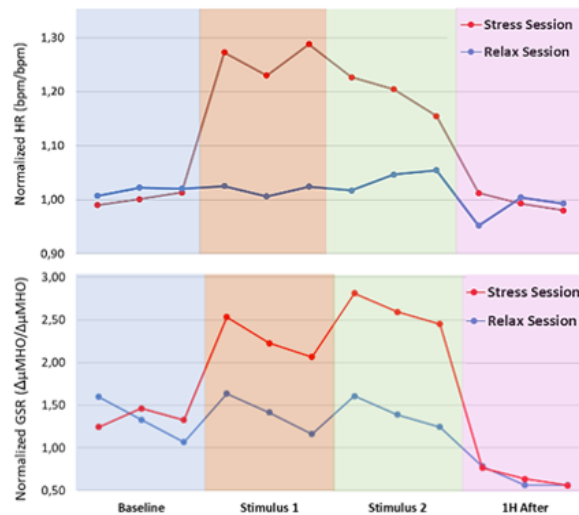


Fig. 1. HR (top) and GSR (bottom) mean responses of stress (red) and relax (blue) sessions

Average HR observations (within each colored region) show significant differences between sessions for both Stimuli (p -value <0.05). However, for GSR, no significant differences were observed between sessions.

High variations in HR and GSR presented under stressor application (versus baseline), validate PASAT suitability to cause physiological stress among volunteers.

4.3 NASA-TLX Response Score

NASA-TLX response scores range from 0 to 100. For RS and SS, NASA-TLX scores were statistically different (13.9 ± 8.5 and 70.4 ± 14.9 , respectively). SS showed a greater range (42.3 to 94.0) when compared with RS (5.0 to 28.0). NASA-TLX response scores are consistent with the results of HR and GSR, thus confirming psychological stress from the PASAT interventions experienced among volunteers.

4.4 VOC Data and Multi-Variate Analysis

GC-IMS data was also processed following previously described workflow [17]. For that purpose, Laboratory Analytical Viewer (LAV) software from GAS Dortmund GmbH, was used to export numerical information of relative intensities within each defined area set of spectra. Each area set included a single peak which represents a unique VOC found in breath. A breath matrix was constructed with 68 (variables) intensity peaks (or breath compounds) against the participations (observations) to perform a multi-variate analysis (MVA)[18]. MVA was conducted using IBM SPSS Statistics for Windows, version 23 (IBM Corp., Armonk, N.Y., USA).

Partial least-squared discriminant analysis (PLS-DA) was initially performed on the post intervention samples (BS02, BS03, BS04 and BS05) from RS and SS. All variables were assigned to a single block and a weighting of $1/\sqrt{\text{Block}}$ was applied producing a total variance equal to 1. Pareto Variance was also selected for base scaling of the data set [18]. The S-plot indicated six possible VOC variables (α_1 to α_6) that changed in response to interventions. Principal component analysis (PCA) for these six VOC variables enabled the separation between the RS and SS observations. Observations were compared at the same measuring point (*e.g.* Stimulus 1 (RS) vs Stimulus 1 (SS)). The score plots obtained from Stimulus 1 and 2 (BS02 and BS03, respectively) are shown in Fig. 2. Responses from SS and RS are seen to cluster and a distinction between breath profiles obtained under the two experimental sessions can be observed.

Principal components (PC) exhibited 50.4% [PC1], 24.9% [PC2] and 16.7% [PC3] of the total explained variance of the data set for the observations after the first stimulus (BS02). Additionally, the results provided 61.5% of sensitivity and 71.4% of specificity for the stressed cases. The observations obtained from samples acquired immediately after the second stimulus (BS03) produced a stronger model and yielded a sensitivity of 78.5% and a selectivity of 71.5%. The total variance modelled by each component was 50.6%, 24.2% and 17.5% for [PC1], [PC2] and [PC3], respectively.

However, PCA for the observations related to the samples acquired 5 min (BS04) and 1 hour (BS05) after the second stimulus showed no distinguishable specific patterns between observations from both sessions. Therefore, no mid and long-term effects of

stress were evident in the collected breath samples. Results of multi-variate analysis of chromatographic data show an immediate response of the breath composition to stress stimuli, which is consistent with HR and GSR responses (Fig. 1).

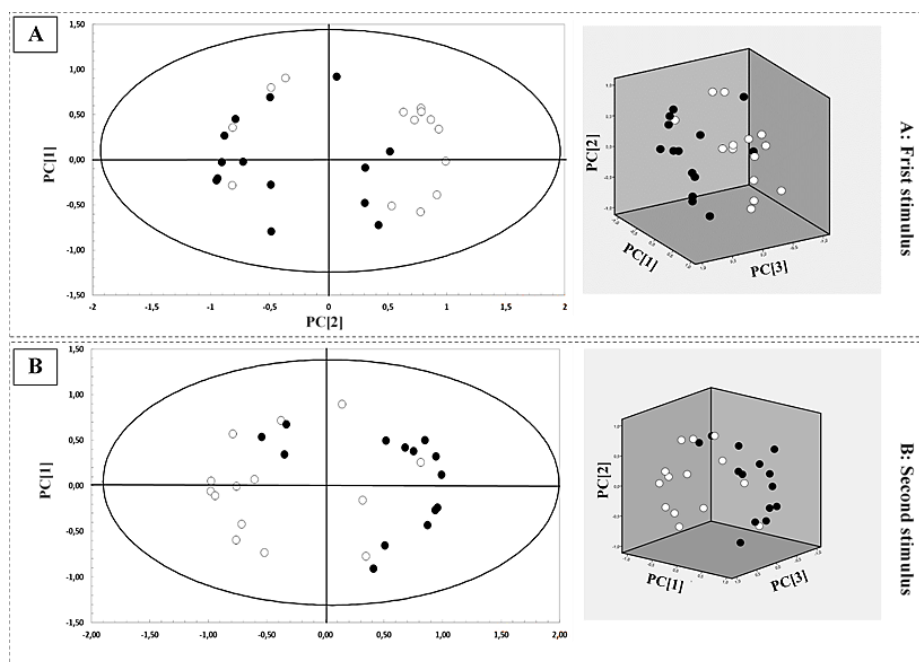


Fig. 2. Unsupervised PCA for the 6 stress sensitive breath compounds identified from PLS-DA. A – First stimulus (BS02): SS (black dots) and RS (white dots) observations. B – Second stimulus (BS03): SS (black dots) and RS (white dots) observations. Observations are presented with the two (left) and three (right) most representative principal components.

4.5 Identification of Discriminant VOCs

IMS spectra provides an increased intensity signal (intensity peak) with a drift time specific for each compound but, in some cases a single compound can produce two or even three peaks (dimers, trimers, etc.). Such effect is related to ionization phenomenon, sample humidity and proton transfer reaction that occurs in the ionization region. Combining IMS capabilities with GC, an additional dimension (retention time) increases the selectivity for VOC detection and quantification in complex biological mixtures.

Table 1 presents the retention and drift times for each of the six-discriminant stress sensitive breath compounds. Those values are specific to certain VOCs. $\alpha 2$, $\alpha 3$ and $\alpha 4$ were identified by cross-checking retention times and drift times in database created from measurements of the corresponding pure compounds (ethanol, 2-propanol and 1-propanol respectively). Nevertheless, not all breath compounds could be identified with

precision. For these cases a list of possible compounds was obtained by consulting a database provided by GAS Dortmund GmbH.

Table 1. Stress sensitive data (retention and drift time) with possible VOC identification.

Peak ID	Drift Time (RIP Rel.)	Retention Time (s)	VOC (Molecular Weight)
$\alpha 1$	1.079	66.780	Styrene (104,15 g/mol)
			1 2-dimethylbenzene (106,17 g/mol)
$\alpha 2$	1.059	24.045	Ethanol (46,07 g/mol)
$\alpha 3$	1.258	25.095	2-propanol (60,10 g/mol)
$\alpha 4$	1.215	28.245	(dimer) 1-propanol (60,09 g/mol)
$\alpha 5$	1.216	28.820	(dimer) 2-propanol (60.10 g/mol)
			2,3-pentanedione (100.12 g/mol)
			Benzaldehyde (106.12 g/mol)
$\alpha 6$	1.124	29.610	(monomer) 1-propanol (60.09 g/mol)
			Ethyl acetate (88.11 g/mol)

5 Conclusions & Future Work

Since stressing situations cause faster breathing (with increased pulse rates and blood-pressure), changes in the VOC profile of breath were expected. Inducing psychological stress (through PASAT interventions) confirmed our assumptions as preliminary results indicate three (of six) identified stress responsive VOCs in human breath. These VOCs were downregulated and may have been depleted through increased ventilation, HR, and respiratory rate during stress. Nevertheless, the combination of the six most discriminant stress responsive compounds was still able to classify stress states with higher sensitivity for breath samples acquired immediately after the most intense stimulus (second). Individuals who were not correctly classified by breath analysis exhibited stress during neutral sessions (induced by the unfamiliar surroundings) or might not have experienced a high stress load.

Demonstrations of the applications of breath analysis in providing next generation health assessment approaches continue to be reported [9]. Physical disease may not be the only contributing factor for breath profile changes and, people undergoing traumatic and emotionally challenging diagnoses related with serious diseases may need to consider such factor. This study shows that human VOC profile is also sensitive to non-physical stimuli. It is expected that the findings herein presented could lead to a quick and non-invasive test to detect mental stress states. However, is still soon to postulate the biological origins and the roles of the present VOCs as part of a stress-sensitive response. The study still needs to be scaled-up and include a higher sample size, with a wider age range and in conditions closer to real-world settings.

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