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# Removal of interfering gases in breath biomarker measurements

A. M. BRATU, C. POPA, C. MATEI, S. BANITA, D. C. A. DUTU, D. C. DUMITRAS*

*Department of Lasers, National Institute for Laser, Plasma and Radiation Physics, 409 Atomistilor St., PO Box MG-36, 077125, Bucharest, ROMANIA*

The analysis of exhaled breath has been proposed as a convenient and safe method for the routine monitoring of metabolic disorders. Exhaled breath may contain a variety of trace amounts of volatile organic compounds (VOCs): acetone, ethanol, ethylene, isoprene, methanol, pentane, etc. and several small inorganic molecules such as ammonia, nitric oxide, carbon oxide, carbon dioxide, etc. Many of these biomarkers have been detected using $CO_2$ laser photoacoustic spectroscopy (LPAS). The measurement of the exhaled volatile hydrocarbons, such as ethylene ($C_2H_4$), a marker of oxidative stress in the human body, is a good noninvasive method to monitor lipid peroxidation. When low concentrations of ethylene have to be measured, it is necessary to remove $CO_2$ (representing ~ 4% in exhaled breath sample) using a potassium hydroxide (KOH) scrubber. We investigated the efficiency of the KOH scrubber using four recipients with different volumes, and we found out that the concentration of $CO_2$ in exhaled air can be reduced significantly only if a certain volume of scrubber is used.

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## 1. Introduction

The analysis of exhaled breath using LPAS method technique is noninvasive and can be performed easily allowing a large number of patients to be studied. Most studies are focused on the detection of a single disease marker as ethylene or ammonia, present in human breath. Ethylene as a biomarker of lipid peroxidation in humans can be detected in the exhaled breath, giving information on various processes occurring inside the body. $CO_2$ laser photoacoustic spectroscopy applied to trace gas analysis exploits the infrared absorption characteristics of gases for detection and measurement [1-3].

The breath air is a mixture of nitrogen, oxygen, carbon dioxide, water, inert gases, and traces of VOCs (Table 1). The matrix elements in breath air vary widely from person to person, both qualitatively and quantitatively, particularly for VOCs. More than 1,000 trace VOCs have been distinguished in human breath air, at concentrations from ppmV (parts per million by volume) to pptV (parts per trillion by volume) levels [4, 5]. Only a small number of VOCs are common to everyone, including isoprene, acetone, ethane, and methanol, which are products of core metabolic processes. In addition to these VOCs, exhaled NO, $H_2$, $NH_3$, and CO are related to health condition and can reflect a potential disease of the individual or a recent exposure to a drug or an environmental pollutant.

*Table 1. Concentration of different components in inhaled and exhaled air.*

| Component | Inhaled air (%) | Exhaled air (%) |
|---|---|---|
| Nitrogen | 78.0 | 78.0 |
| Oxygen | 21.0 | 16.0 |
| Carbon dioxide | 0.04 | 3.0-5.0 |
| Argon | 0.93 | 1.0 |
| Water | 2.0 | 5.0-6.0 |
| Other<br>- ammonia<br>- ethylene | 0.01 | <br>$25 \times 10^{-6}$ (250 ppb)<br>$3 \times 10^{-6}$ (30 ppb) |

The exhaled air is a heterogeneous gas. For a healthy individual, the first part of a exhaled breath, roughly 150 mL, consists of “dead-space” air from the upper airways (such as the mouth and trachea), where air does not come into contact with the alveoli of the lungs. The following part of a breath, about 350 mL, is "alveolar" breath, which comes from the lungs, where gaseous exchange between the blood and breath air takes place. Dead space air can be interpreted as essential for the detection and depends on the type of molecule detected from the breath test. For example, the dead-space is used to quantify the amount of the NO molecules. In the case of an asthmatic patient, if the airways are inflamed, a high-level of NO is released into the airways and into the dead-space air. But for VOCs exchanged between blood and alveolar air, the dead-space air is a “contaminant” diluting the concentrations of VOCs when breath air is collected. In terms of the origin of the collected breath gases, there are three basic collection approaches: 1. *upper airway collection* for NO test; this means that only dead-space gas is collected (it is only for the NO test); 2. *alveolar collection*; this means that pure alveolar gas is collected (for tests of other inorganic gases and VOCs); 3. *mixed expiratory collection*; this means that total breath air, including dead-space air and alveolar gas is collected (appropriate for tests of special gases and VOCs). Because the mixed expiratory collection method is easy to perform in spontaneously breathing subjects requiring no additional equipment, it has been most frequently used in practical applications. However, concentrations of endogenous substances in alveolar air are two to three times higher than those found in mixed expiratory samples, because there is no dilution by dead-space gas.

A healthy adult human has a respiratory rate of 12-15 breaths/min at rest, inspiring and expiring 6-8 L of air per minute. $O_2$ enters the blood and $CO_2$ is eliminated through the alveoli. When the end-tidal concentration of $CO_2$ in healthy persons is measured, a large change of $CO_2$ concentration is observed between the inhaled air (~ 0.04%) and the exhaled air (~ 4%) [6]. The exact amount of exhaled $CO_2$ varies according to the fitness, energy expenditure and diet of a particular person, with regular values of 3-5%. Due to this high concentration of carbon dioxide in the breath and because $CO_2$ laser lines are absorbed by this gas, it is necessary to remove most of the carbon dioxide from the exhaled air by introducing a scrubber filled with a chemical active agent, KOH in our case [7, 8].

We have investigated the efficiency of the KOH scrubber using four recipients with different volumes (13 $cm^3$, 45$cm^3$, 120 $cm^3$, and 213 $cm^3$, respectively), and we found out what type has to be used in order to reduce efficiently the amount of $CO_2$ from the exhaled air sample.

## 2. Gas interference

Interference of other absorbing substances may impair the theoretical detection limit in a multicomponent analysis of the real samples. Such interference may be caused by other molecular systems present in the environment or substances that are entrained by the carrier flux. If an interfering species is present in the environment, its effect can be minimized by either the introduction of scrubbers and cryogenic traps or the use of dual beam techniques using two photoacoustic (PA) cells.

The $CO_2$ laser spectral outputs occur in the wavelength region where a large number of compounds possess strong absorption features and where absorptive interferences from water vapors, carbon dioxide, and other major atmospheric gaseous components may influence the measurements.

Due to the exact coincidence of the $CO_2$ vibrational-rotational transitions with the $CO_2$ laser lines, the carbon dioxide at high concentration in comparison with trace gases like $C_2H_4$ is inevitably excited by $CO_2$ laser radiation and the related photoacoustic signal may exceed the trace signal by many orders of magnitude. The absorption coefficient increases strongly with temperature, but it is independent of the $CO_2$ concentration over a wide range. Ethylene can be excited by the 10P(14) line of the $CO_2$ laser, where the maximum absorption coefficient $\alpha(C_2H_4)$ has a value of 30.4 $cm^{-1}$ $atm^{-1}$ [1] and ammonia by the 9R(30) line, where $\alpha(NH_3)$ = 56 $cm^{-1}$ $atm^{-1}$ [2]. A 4% concentration of $CO_2$ has an absorption strength comparable to 2760 ppbV of $C_2H_4$ (at the 10P(14) laser line, $\alpha(CO_2) = 2.1x10^{-3}$ $atm^{-1}cm^{-1}$ and $c(C_2H_4) = c(CO_2)\alpha(CO_2)/\alpha(C_2H_4)$. This equivalent ethylene concentration was found also experimentally (see Fig. 4). So, the photoacoustic signal is 100 times higher owing to exhaled carbon dioxide in comparison with the usual concentration of ethylene in exhaled air. Similarly, at the 9R(30) line of $CO_2$, the same concentration of $CO_2$ has an absorption coefficient equal to that of 1500 ppbV of $NH_3$. This value is also considerably higher (6 times) compared to the real range of breath concentration which is situated approximately at 250 ppb for ammonia.

Water vapor exhibits a broad continuum with occasional weak lines in the frequency range of the $CO_2$ laser (for $H_2O$ at the 10P (14) laser line, $\alpha(H_2O)$ = $2.85x10^{-5}$ $atm^{-1}cm^{-1}$). The two dominant peaks are the absorption lines on 10R(20) and the most favorable one for ambient air measurement, the 10P(40) laser transition. A 5% concentration of $H_2O$ has an absorption strength comparable to 46.9 ppbV of $C_2H_4$, that is the normal concentration of water in exhaled air has approximately the same influence in the photoacoustic signal as the normal concentration of ethylene.

Due to the additive character of the photoacoustic signal under normal pressure conditions, the presence of a large amount of water vapor and carbon dioxide impedes $C_2H_4$ detection in the low-concentration range (ppbV). Consequently, some means of selective spectral discrimination is required if ethylene is to be detected interference free in the matrix of absorbing gases. There are several ways to overcome this problem. One way is to remove $CO_2$ from the flowing sample by absorption on a KOH-based scrubber inserted between the sampling cell and the PA cell. Taking into account the nature of the specific chemical reactions involved in the $CO_2$ removal by KOH, a certain amount of water is also absorbed from the sample passing the scrubber. In this way, concentrations below 1 ppmV $CO_2$ (equivalent to a concentration of 0.07 ppbV of $C_2H_4$) can be achieved without influencing the $C_2H_4$ or $NH_3$ concentration.

## 3. Experimental setup

$CO_2$ laser photoacoustic spectroscopy offers a sensitive technique for detection and monitoring of trace gases at low concentrations. The block diagram of the laser photoacoustic spectrometer (described in details elsewhere [1]) is presented in Fig. 1.

The CW, tunable $CO_2$-laser beam is chopped, focused by a ZnSe lens, and introduced in the PA cell. After passage through the PA cell, the power of the laser beam is measured by a laser powermeter with a measuring head.

Its digital output is introduced in the data acquisition interface module together with the output from the lock-in amplifier. All experimental data are processed in real time and stored by a computer. The trace gas concentration is proportional to the photoacoustic signal to laser power ratio and is inverse proportional to gas absorption coefficient and cell responsivity.

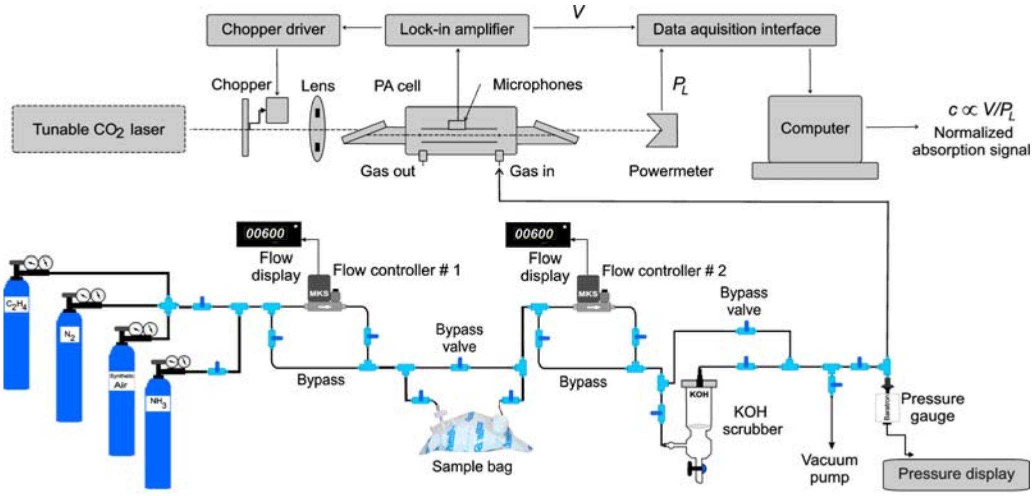

*Fig. 1. General scheme of the photoacoustic detection system with a $CO_2$ laser and the gas handling station.*

To calibrate the measurement in absolute values of pressure, it is necessary to know the gas absorption coefficient at a given laser wavelength and the cell responsivity. The last one was determined experimentally by using a reference gas mixture (ethylene in nitrogen). The cell responsivity is the signal per unit power per unit absorption coefficient; in our case, the signal per unit power is 11.6 mV/4.0 W = $2.9\text{x}10^{-3}$ V/W (rms value) or $8.2\text{x}10^{-3}$ V/W (peak-to-peak value) for 0.96 ppmV of $C_2H_4$ (the absorption coefficient $\alpha^* = 30.4$ $\text{cm}^{-1}$ $\text{atm}^{-1}$ x $0.96\text{x}10^{-6}$ atm = $2.92\text{x}10^{-5}$ $\text{cm}^{-1}$, where $\alpha = 30.4$ $\text{cm}^{-1}$ $\text{atm}^{-1}$ is the absorption coefficient of $C_2H_4$ at 10P(14) line of the $CO_2$ laser), so that $R = 8.2\text{x}10^{-3}$ V/W/$2.92\text{x}10^{-5}$ $\text{cm}^{-1} \cong 280$ V cm/W; the same responsivity was obtained with the etalon mixture of 10 ppmV of $C_2H_4$ in $N_2$: $R = 8.4\text{x}10^{-2}$ V/W/$3\text{x}10^{-4}$ $\text{cm}^{-1} \cong 280$ V cm/W.

It has to be mentioned that the responsivity depends strongly on total pressure in the PA cell. This dependence is shown in Fig. 2.

The gas handling system is an important part of the experimental set-up for the trace gas concentration measurements, ensuring gas purity in the PA cell (Fig. 1). The handling system can be used to introduce the sample gas in the PA cell at a controlled flow rate, to pump out the cell, and to monitor the total pressure of gas mixtures. The gas handling system includes two gas flow controllers MKS 1179A (0–1000 sccm) [sccm - standard cubic centimeters per minute, 1 sccm at $0^oC$ = $7.436\text{x}10^{-7}$ mol/sec] and MKS 2259CC (0–200 sccm), which are connected to a digital four-channel instrument MKS 247C.

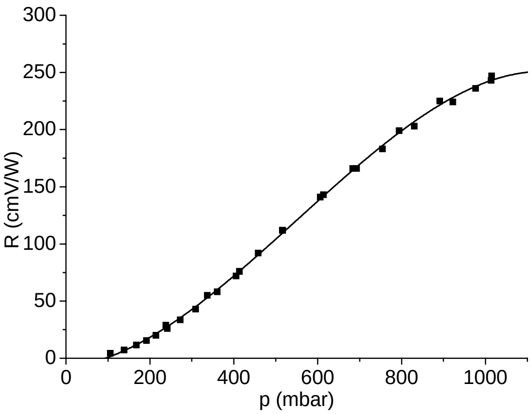

*Fig. 2. Dependence of the PA cell responsivity on the total gas pressure in the cell (measured with 1 ppmV ethylene in nitrogen).*

The breath samples were collected in 0.75-liter aluminum-coated bags equipped with valves that sealed them after filling. The stored breath samples are transferred from bags into the measuring cell by the gas flow controller #2 (MKS 1179A).

Before entering the photoacoustic cell, the gas mixture passes through a KOH scrubber (Fig. 3), which retains most of the interfering carbon dioxide. The removal of $CO_2$ is limited to the absorbent surface of the pellets. Hence, the larger the surface area or the more porous the granular solid, the larger the capacity of the system to absorb $CO_2$. At the same time, the flow resistance varies inversely proportional to the particle size. Large particles offer less resistance, but have the disadvantage of providing a smaller total area for reaction. The granules of KOH that we used were typically Merck KOH pellets GR for analysis, ovals with approximate dimensions of 10x7x2 mm$^3$. When residence time (time of contact between $CO_2$ and absorbent) is less than 1 second, $CO_2$ absorption capacity is greatly reduced, so we introduced flow controllers in order to ensure this pre-requisite.

Potassiun hydroxide is a caustic compound of strong alkaline chemical, dissolving readily in water, giving off much heat and forming a caustic solution. It is a white deliquescent solid in the form of pellets obtained by concentration of purified electrolytic potassium hydroxide solution with very low chloride content. It reacts violently with acid and it is corrosive in moist air toward metals such as zinc, aluminum, tin and lead forming a combustible, explosive gas. It absorbs rapidly carbon dioxide and water from air [9]. Cautions must be taken when used because the inhaled dust is caustic and irritant, and touching skin or clothes could lead to less or more severe chemical burnings.

*Fig. 3. KOH scrubber and aluminum-coated bags.*

The transfer of the sample gas from the aluminum bag to the PA cell was realized at a controlled flow rates of 300 sccm or 600 sccm, and the pressure of the gases introduced in the PA cell was determined with a Baratron pressure gauge. In this way, the transfer time (the time necessary the sample gas to pass through KOH scrubber) is increased to 7 min for a flow rate of 600 sccm, or to 15 min for a flow rate of 300 sccm. The final pressure inside the PA cell, measured for the breath samples from healthy humans, attains usually ~ 700 mbar (this pressure is the result of the initial pressure in the sample bag and the volumes of the bag and the PA cell, respectively). Taking into account the fact that the pressure in sample bags filled from healthy humans and from suffering patients differ from case to case, it is necessary to correct the cell responsivity at the pressure inside the PA cell for each measurement, according to Fig. 2.

## 4. Results and discussion

Exhaled human breath contains $CO_2$ in much higher quantities than other compounds and interferes with the useful signal due to ethylene absorption. In order to reduce the high quantity of $CO_2$ from human breath, it is necessary to introduce in the system a chemical scrubber such as KOH [10-12]. The KOH scrubber must neither change the ethylene concentration level, nor introduce new interfering gases. We investigated the efficiency of the KOH scrubber using three recipients with different volumes (13 cm$^3$, 45 cm$^3$, 120 cm$^3$, 213 cm$^3$), and found out what type has to be used in order to reduce efficiently the amount of the $CO_2$ from the exhaled air sample.

The measurements were made each time on the same person (healthy female, 30 years old) and with a new filling of KOH pellets. The gas from the sample bag was transferred into the PA cell at a controlled flow rate of 300 sccm (only for the 13 cm$^3$ trap) or 600 sccm, in order to ensure a sufficient time of flow in the scrubber column and to minimize any tendency for the vapor to stick to the cell walls or any other effects of internal outgassing of contaminants, which would otherwise lead to increase background signals during an experimental run. The typical resulting final pressure inside the PA cell was 700 mbar and the corresponding responsivity was 170 cmV/W.

The experimental results without the KOH scrubber showed an equivalent ethylene absorption concentration of 2750 ppbV (with alveolar air collection) and 2350 ppbV (with mixed expiratory air collection), representing mainly the contribution of ethylene, carbon dioxide, water vapors and ammonia to the absorption of 10P(14) $CO_2$ laser line (Fig. 4). We tested the efficiency of traps filled with KOH and having different volumes (between 13 cm$^3$ and 213 cm$^3$) in removing $CO_2$ from exhaled air.

For the first measurement we used a trap with a small volume of 13 cm$^3$ of KOH scrubber, and we obtained a decrease of the PA signal down to 1-3 mV. The equivalent ethylene concentration was 435 ppb and 240 ppb, respectively (alveolar air collection vs. mixed expiratory air collection), indicating that the $CO_2$ concentration was reduced by factors of 6.3 and 9.8, respectively. Only in the case of this trap we observed a peculiar behavior. Even if the laser power is constant, the PA signal and consequently the equivalent ethylene concentration increases in time after transferring the gas sample in PA cell. The increase of concentration starts from 50 ppbV and continues until it stabilizes at a level of 435/240 ppbV (after 10-15 minutes). It is known that $C_2H_4$ (28.05 g/mol molar mass) is lighter than $CO_2$ (44.0099 g/mol molar mass). Because of that, we can say that after passing the KOH scrubber, first $C_2H_4$ enters in the PA cell and then $CO_2$ when the trap is no longer effective. So, at the beginning, we measured only the $C_2H_4$ concentration and then $CO_2$ starts to strongly interfere in absorption. It is possible that due to the geometry of the cell, a longer time is required in order to attain the total homogeneity of the molecules inside the resonant tube of the cell, but this is not advantageous for repeated measurements.

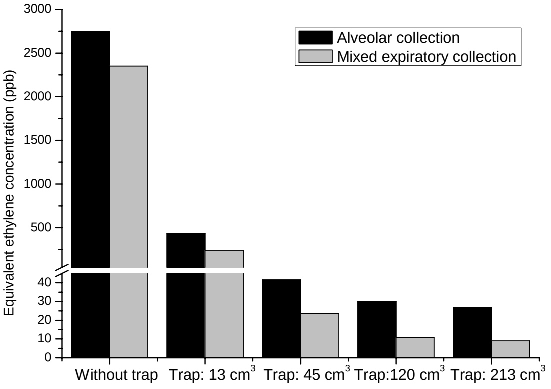

*Fig. 4. Efficiency of KOH traps for $CO_2$ removal from exhaled air.*

Larger KOH traps proved to be more efficient in removal of $CO_2$ from the exhaled air. For the traps with volumes of 45 $cm^3$, 120 $cm^3$, and 213 $cm^3$, respectively, the measured equivalent ethylene concentrations were 41.5/23.6 ppb, 30/10.8 ppb, and 26.8/9.1 ppb, respectively.

For larger traps (120 $cm^3$ and 213 $cm^3$), approximately same results were obtained, indicating that most of the $CO_2$ was removed. By using larger traps, a higher transfer rate of the gas mixture in the PA cell is possible, doubling the flow rate to 600 sccm.

For the two largest volumes, we succeeded to reduce the $CO_2$ content from the exhaled air at a level influencing no more the $C_2H_4$ and $NH_3$ concentration values, fact proved by the constant evolution in time of all parameters. Therefore, the trap is effective only for a large amount of KOH pellets. We found that a minimum volume of 120 $cm^3$ of KOH scrubber and a transfer rate of 600 sccm were optimum to insure the required efficiency.

We measured also the efficiency of the KOH scrubber when it is used for multiple measurements (Fig. 5). A clear saturation effect is evident: the KOH scrubber is not anymore efficient when the same fill is used for multiple runs (it cannot absorb completely the $CO_2$ from the gas mixture). In the case of alveolar collection, the equivalent ethylene concentration increases by 2.3 times for the second run, by 2.6 times for the third run and by 3.4 times for the fourth run. When we measured the mixed expiratory collection, this saturation effect is even larger: the equivalent ethylene concentration increases by 2.4 times for the second run, by 8.5 times for the third run and by 20.2 times for the fourth run.

The lungs and airways are always moist, and inspired gas is rapidly saturated with water vapor in the upper segments of the respiratory system. The temperature in the airways and lungs is most identical with deep body temperature (approximately $37^0$C); at this temperature water vapor has a partial pressure of 47 torr (~ 6.2%). The increased saturation found at the third and fourth run for mixed expiratory collection is explained by a higher quantity of water vapors in exhaled breath (originating both from lungs and from upper segments of the respiratory system).

The conclusion is that a new fill of KOH scrubber must be introduced after each measurement.

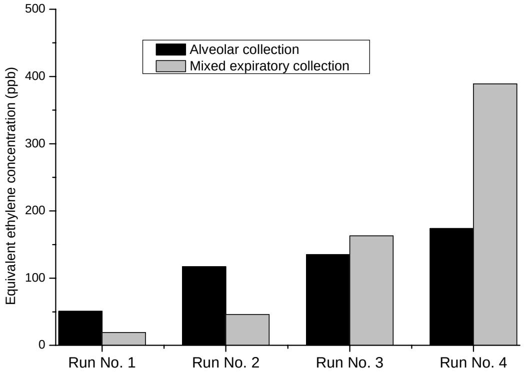

*Fig. 5. Decrease of KOH trap efficiency when the same fill was used for multiple measurements.*

Analyzing the four cases when we inserted the scrubber, the dependence between the removed content of $CO_2$ and the used KOH quantity proved to be nonlinear, as one could expect. If we consider the content of the sample totally free of $CO_2$ after passing through the 213 $cm^3$ and 120 $cm^3$ KOH traps, we calculated a residual content of $CO_2$ in alveolar collection of 0.58% (5800 ppm) for the 13 $cm^3$ trap and of 0.016% (160 ppm) for the 45 $cm^3$ trap (less than half of the $CO_2$ concentration in the inhaled air).

The nonlinearity of the $CO_2$ removal could be explained by the mechanism of the chemical reactions. First, the $CO_2$ combines with the water vapors present in the exhaled air in the form of carbonic acid:

$$CO_2 + H_2O \Rightarrow H_2CO_3. \qquad (1)$$

Further, the last one combines with the KOH, creating potassium carbonate and water, and releasing a small amount of heat:

$$H_2CO_3 + 2KOH \Rightarrow K_2CO_3 + 2H_2O + \text{Energy} \qquad (2)$$

In the same time, $K_2CO_3$ is a highly hygroscopic compound with a retaining capacity of 0.2 g $H_2O$/1 g $K_2CO_3$, so the generated water will be only partially returned in the circuit.

The water is of high importance in limiting the rate of $CO_2$ absorption. High $CO_2$ concentrations entering the KOH absorber generates large quantities of water, because the reaction (2) is generating water. We know that the absorption rate is greater thanks to the film of moisture coating the pellets, but the same film impedes the access to the active potassium hydroxide pellet volume. More dedicated studies should be made in order to establish the moisture content for an optimum rate of absorption.

## 5. Conclusions

The present work was carried out by applying a methodology which assured better conditions to measure real concentrations of ethylene and ammonia in the exhaled breath.

We determined experimentally that in the process of $CO_2$ removal from the breath air samples, a quantity of minimum 120 $cm^3$ KOH pellets should be used for a sampling bag of 750 mL in order to keep the detection of ethylene and ammonia traces free of $CO_2$ interference. It should be mentioned that this volume of 120 $cm^3$ must be reconsidered for samples with a greater volume (> 750 mL) or in conditions of increasing the gas flow rate.

By this investigation, with a high degree of accuracy, we proved that the photoacoustic technique could measure trace amounts of ethylene and ammonia in the exhaled air from patients with damages caused by free radicals and patients with renal failure, by using the $CO_2$ laser lines where the ethylene [10P(14)] and ammonia [9R(30)] absorption coefficients have the maximum values [13, 14].

Noninvasive medical diagnosis using breath analysis method is a topic of great interest because of its ability to distinguish more than 200 compounds in human breath. Many of these compounds, if measured accurately at very low concentration levels, typically in the range of few ppbV, can be used to identify particular medical conditions.

With the relevant characteristics of high sensitivity and specificity, laser photoacoustic spectroscopy holds a great potential for medical diagnostics.

## References

[1] D.C. Dumitras, D.C. Dutu, C. Matei, A. Magureanu, M. Petrus, C. Popa, J. Optoelectron. Adv. Mater. **9**, 3655 (2007).

[2] D.C. Dumitras, S. Banita, A. M. Bratu, R. Cernat, D. C. A. Dutu, C. Matei, M. Patachia, M. Petrus, C. Popa, Infrared Phys. Technol. **53**, 308 (2010).

[3] D.C. Dumitras, D.C. Dutu, C. Matei, R. Cernat, S. Banita, M. Patachia, A.M. Bratu, M. Petrus, C. Popa, Laser Phys. **21**, 796 (2011).

[4] W. Ca, Y. Duan, Clinical Chemistry **52**, 800 (2006).

[5] C. Wang, P. Sahay, Sensors **9**, 8230 (2009).

[6] M. Folke, L Cernerud, M Ekstrom, B Hok, Med. Biol. Eng. Comput. **41**, 377 (2003).

[7] R.Cernat, C. Matei, A.M. Bratu, C. Popa, D.C.A. Dutu, M. Patachia, M. Petrus, S. Banita, D.C. Dumitras, Rom. Rep. Phys **62**, 610 (2010).

[8] F.J.M. Harren, F.G.C.Bijnen, J. Reuss, L.A.C.J. Voesenek, C.W.P.M. Blom, Appl. Phys. B **50**, 137 (1990).

[9] http://chemicalland21.com/industrialchem/inorganic/koh.htm.

[10] T.H. Risby, S.F. Solga, Appl. Phys. B **85**, 421 (2006).

[11] L.A.C.J Voesenek, M. Banga, J.H.G.M. Rijnders, E.J.W. Visser, F.J.M. Harren, R.W. Brailsford, M. B. Jackson, C.W.P.M. Blom, Annals of Botany **76** (Supplement A), 57 (1997).

[12] E. V. Stepanov, Physics of Wave Phenomena **15**, 149 (2007).

[13] C. Popa, A.M. Bratu, C. Matei, R. Cernat, A. Popescu, D.C. Dumitras, Laser Phys. **21**, 1336 (2011).

[14] C. Popa, R. Cernat, D.C.A. Duțu, D.C. Dumitras, Appl. Phys. B 103, (2011).

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[*] Corresponding author: dan.dumitras@inflpr.ro