The Performance of CO₂ Laser Photoacoustic Spectrometer In Concentration Acetone Detection As Biomarker For Diabetes Mellitus Type 2

F H Tyas, J G Nikita, D K Apriyanto, Mitrayana* and M N Amin
Department of Physics, Universitas Gadjah Mada, Yogyakarta, INDONESIA

*Corresponding author: mitrayana@ugm.ac.id

Abstract. Breath analysis is useful for the diagnosis of human diseases and monitoring of metabolic status. However, because of the low concentrations and the large numbers of compounds in the breath, the breath analysis requires highly sensitive and highly selective instruments to identify and determine the concentrations of certain biomarkers [1]. Various methods developed over the past 20 years to detect biomarker gases [2]. CO₂ laser photoacoustic spectroscopy offers a sensitive technique for the detection and monitoring of gas footprints at low concentrations [3]. The performance of photoacoustic spectrometer (PAS) examined with intracavity configuration. In this research, the highest observed intracavity power was (49,96 ± 0,02) W for active medium gas composition He: N₂: CO₂ at 30:50:50. The highest laser absorption line for standard acetone gas set at 10P20, and the lowest detection limit set at (30 ± 4) ppb. For application purposes, the photoacoustic spectrometer was used to measure the concentration of acetone gas in exhaled gases from a group of patients with type 2 diabetes mellitus and a group of healthy volunteers. Exhaled gas sampling method took manually, and the measurement result was examined using multicomponent analysis. The measurement showed that the highest acetone gas concentration for type 2 diabetes mellitus patients was (162 ± 3) × 10 ppb and the lowest one was (101 ± 3) × 10 ppb. Furthermore, for healthy volunteers, the highest acetone gas concentration was (85 ± 3) × 10 ppb and the lowest one was (15 ± 3) × 10 ppb.

Keywords: CO₂ laser photoacoustic spectrometer, acetone, diabetes mellitus, multicomponent analysis

1. Introduction

Breath analysis is useful for the diagnosis of human diseases and monitoring of metabolic status. However, the breath analysis requires highly sensitive and highly selective instruments to identify and determine the concentrations of certain biomarkers [1]. CO₂ laser photoacoustic spectroscopy offers a sensitive technique for the detection and monitoring of gas footprints at low concentrations. Because it ensures high output power in the wavelength region of 9 – 11μm where more than 250 gas molecules exhibit strong absorption of bands in atmospheric, industrial, medical, military and science environments [3]. CO₂ lasers have extensive tunability and high laser power that allows measurement of mixtures of several gases at concentrations (sub) parts per billion volume, 1 ppb = 1: 10⁹ [4].

Results from Basic Health Research Reports stated that the prevalence of DM patients in Indonesia in 2013 (2.1%) is increased compared to 2007 (1.1%). The highest prevalence of DM found in Yogyakarta province with 2.6% prevalence followed by Jakarta with 2.5% and Sulawesi 2.4% [5]. This study examines the measurement of acetone gas concentration as a biological marker (biomarker)
of type 2 diabetes mellitus through human breath. Type 2 diabetes mellitus is selected because, in adult age range, type 2 diabetes accounts for about 90% to 95% of all diagnosed diabetic cases [6]. The photoacoustic spectroscopy method of the CO$_2$ laser was applied in this study to measure the concentration of acetone gas (C$_3$H$_6$O) in the breath sample. This study conducted on healthy people volunteers and patients with type 2 diabetes mellitus who has a range of ages 25-80 years, amounting to 31 individuals each.

2. Experiment details
The research materials used in this research were exhalation gas samples of DSH patients and healthy volunteers with age range 25 - 80 years which consist of 31 people in each group. CO$_2$ gas, N$_2$, and He as the active medium of the CO$_2$ laser. Distilled water as a coolant for the CO$_2$ laser tube. N$_2$ gas to clean the FA cells and determine the background signal. The standard gas (ethylene, ammonia, and acetone) as a calibrator. KOH, and CaCl$_2$ to fill the scrubber tube and 75% alcohol for cleaning the hose covering device on the sample bag.

Equipment calibration was also carried out on three standard gases of ethylene, acetone and ammonia gas to obtain the relationship matrix of gas concentration to the selected laser line, 10P14 for ethylene, 10P20 for acetone and 10R14 for ammonia. In this research, CO$_2$ laser power optimization is done to get optimal laser output power. Some of the factors that affect the optimization of are: the ratio of the active ingredient laser (CO$_2$ gas, N$_2$, and He), the discharge arrangements in the form of variations of the power supply and voltage current and grating position settings.

In this study, every patient and healthy person volunteers are guided to take a deep breath; then breath is retained and then streamed to a hose connected to scrubbers and sample bag. The scrubber contains KOH and CaCl$_2$ which serves to filter CO$_2$ and H$_2$O in the volunteers’ exhaled gas. Replacement of KOH and CaCl$_2$ on the scrubber is done periodically as the powders begin to appear solid and the volunteers feel heavy in blowing them. Hoses used for exhaling the gas washed with alcohol before being used by volunteers and hoses replaced for each volunteer. Sample bag that has been used then cleaned by using a vacuum pump and nitrogen gas. After the sample bag cleaned, it can reuse for sampling the next volunteer breath.

The process of measuring breath gas samples of patients with type 2 diabetes in this study was carried out by flowing gas into the sample bag with the flow pump into the photoacoustic cell. A photoacoustic cell is a place of interaction between breath gas with CO$_2$ laser beam radiation in which there is a microphone as a detector of the photoacoustic signal generated from the interaction. In this study, gas samples in photoacoustic cells were measured under conditions of gas flowing systems and recorded on the three laser absorption lines: ethylene (10P14), acetone (10P20) and ammonia (10R14) using LabVIEW software.

Results of signal and power recorded on processed using software LabView origin to obtain the normalized signal. Then put on a normalized signal matrix equation as follows:

$$\begin{bmatrix}
S_1 \\
S_2 \\
S_3
\end{bmatrix} =
\begin{bmatrix}
k_{11} & k_{12} & k_{13} \\
k_{21} & k_{22} & k_{23} \\
k_{31} & k_{32} & k_{33}
\end{bmatrix}
\begin{bmatrix}
C_1 \\
C_2 \\
C_3
\end{bmatrix},$$

with $k_{ij}$ searched through each gradient of gas standards used are ethylene, acetone, and ammonia analyzed on several variations of laser absorption lines are 10P14, 10P14, 10R14 in various concentration.

After each sample of breath was recorded and resulted in a normalized signal recording, data analysis was performed to calculate the amount of biomarker gas concentration in each sample by the inverse multicomponent matrix. In each new bag sample measurement, cell cleansing done by using nitrogen gas that passed on photoacoustic cells.

3. Result and discussion
The composition ratio of the laser’s active medium gas CO\(_2\) (He: N\(_2\): CO\(_2\)) in this study was 30:50:50 with laser’s operating voltage ranging from 8.18 to 9.13 kV and the current ranged from 11.15 to 13.53 mA. The adjustment of the composition of active medium gas generates a maximum intracavity power of (49.96 ± 0.02) W and producing two groups of CO\(_2\) laser 10R and 10P.

The result of CO\(_2\) laser scanning generates the laser line output spectrum with the strongest FA signal of acetone gas is found at line 10P20. The spectrum patterns of acetone absorption have a distinct characteristic than other gas spectrum patterns such as ethylene gases which have the strongest signals only on a single 10P14 laser line and ammonia gas on a single 10R14 laser line. Acetone gas absorption lies in almost all CO\(_2\) laser lines, although it has a small signal.

![Figure 1. The strongest FA signal of acetone gas](image)

In an axial flowing-type CO\(_2\) laser, the turbulence in the active medium gas stream (He, N\(_2\) and CO\(_2\)) simply affects the power output during the sample recording process. However, this problem solved by normalizing the photoacoustic signal to its laser power. Linearity curves for several variations of ammonia and ethylene gas concentrations analyzed by recording the photoacoustic signals on the three strongest absorption lines for ethylene, acetone and ammonia gas, laser lines 10P14, 10P20 and 10R14. The gradient of each linearity curve that obtained is then used as a multi-component matrix of analysis as shown in (2):

\[
\begin{bmatrix}
S_1 \\
S_2 \\
S_3
\end{bmatrix} =
\begin{bmatrix}
0.023 & 0.002 & 0.0015 \\
0.0114 & 0.0179 & 0.0059 \\
0.001 & 0.0002 & 0.0256
\end{bmatrix}
\begin{bmatrix}
C_1 \\
C_2 \\
C_3
\end{bmatrix}.
\]  

(2)

Then to find the concentration of the sample with a known amplitude of the photoacoustic signal, matrix inversion is applied to the equation (2) and resulted respectively being the concentration of ethylene gas, acetone gas and ammonia gas as shown in (3):

\[
\begin{bmatrix}
C_1 \\
C_2 \\
C_3
\end{bmatrix} =
\begin{bmatrix}
46.088 & -5.132 & -1.517 \\
-28.833 & 59.221 & -11.959 \\
-1.575 & -0.262 & 39.215
\end{bmatrix}
\begin{bmatrix}
S_1 \\
S_2 \\
S_3
\end{bmatrix}.
\]  

(3)
With $C_1$, $C_2$, and $C_3$ respectively being the concentration of ethylene gas, the concentration of acetone gas and the concentration of ammonia gas. While $(S/I)_1$, $(S/I)_2$ and $(S/I)_3$ are respectively normalized photoacoustic signals at line 10P14, 10P20 and 10R14.

Figure 2 shows that the concentration of acetone possessed by each DM type 2 patient is higher than that of healthy volunteers. In this research, the highest acetone gas concentration for diabetes mellitus type 2 patients was $(162 \pm 3) \times 10$ ppb and the lowest one was $(101 \pm 3) \times 10$ ppb. Furthermore, for healthy volunteers, the highest acetone gas concentration was $(85 \pm 3) \times 10$ ppb and the lowest one was $(15 \pm 3) \times 10$ ppb. The average value of the concentration of acetone gas of type 2 DM patients $(118 \pm 3) \times 10$ ppb and in healthy volunteers the average concentration of acetone is $(62 \pm 3) \times 10$ ppb.

![Figure 2: The acetone Gas Concentration in Healthy Volunteers' Breath and Patient with DM Type 2’s Breath Measured by Photoacoustic Spectroscopy Laser CO$_2$](image)

4. Conclusion

SFA Optimization The CO$_2$ laser produces two groups of 10R and 10P with maximum intracavity $(49.96 \pm 0.02)$ W and a quality factor on the 10P20 was $(28 \pm 5)$. The SFA CO$_2$ laser demonstrates good performance in the measurement of voluntary volunteer gas DM type 2 patients and healthy person volunteers with the lowest detection limit on acetone line (10P20) was $(30 \pm 4)$ ppb. The highest acetone gas concentration for diabetes mellitus type 2 patients was $(162 \pm 3) \times 10$ ppb and the lowest one was $(101 \pm 3) \times 10$ ppb. Furthermore, for healthy volunteers, the highest acetone gas concentration was $(85 \pm 3) \times 10$ ppb and the lowest one was $(15 \pm 3) \times 10$ ppb. The average value of the concentration of acetone gas of type 2 DM patients $(118 \pm 3) \times 10$ ppb and in healthy volunteers the average concentration of acetone amounted to $(62 \pm 3) \times 10$ ppb.
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