

CHEMICAL CONCERNTRATION CALCULATION BASED ON
SPECTROGRAM

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ABSTRACT

In this project, the chemical concentration is identified based on a spectrogram. In chemistry, concentration is defined as the abundance of a constituent divided by the total volume of a mixture. The term concentration can be applied to any kind of chemical mixture, but most frequently it refers to solutes in homogeneous solutions. Meanwhile, a spectrogram is a time-varying spectral representation (forming an image of a graph) that shows how the spectral density of a signal varies with time. It is an instrument used to measure properties of light over a specific portion of the electromagnetic spectrum, typically used in spectroscopic analysis to identify materials. The purpose of this project was to develop the systems that can identify the concentration of a compound by using a spectrogram. Using MATLAB, the chemical concentration can be calculated based on the compound of the solution. By using Acrylic acid as an example; which is the simplest unsaturated carboxylic acid which has a double bond and a carboxyl group in a 3-carbon molecule with the formula $\text{CH}_2=\text{CHCOOH}$, we can determine the concentration percentages of the compound that form acrylic acid. Based on the graph that is plotted by using MATLAB, the area under the graph is calculated. With the area, we can calculate the concentration directly based on the data from HPLC-high performance liquid chromatography.

ABSTRAK

Dalam projek ini, kepekatan bahan kimia dikenalpasti melalui spektrogram. Dalam bidang kimia, kepekatan ditakrifkan sebagai konstituen yang dibahagikan dengan jumlah keseluruhan campuran yang banyak. Istilah kepekatan boleh digunakan untuk apa-apa jenis campuran kimia, tetapi yang paling kerap ia merujuk kepada bahan larut dalam penyelesaian homogen. Sementara itu, spektrogram mewakili perubahan yang berkaitan dengan masa spektrum (menbentuk graf) yang menunjukkan bagaimana isyarat ketumpatan spektrum berubah dengan masa. Ia merupakan instrumen yang digunakan untuk mengukur sifat cahaya yang mana sebahagian daripada spektrum elektromagnet, biasanya digunakan dalam analisis spektroskopi untuk mengenal pasti bahan-bahan tertentu. Tujuan projek ini adalah untuk membangunkan sistem yang boleh mengenalpasti kepekatan sebatian dengan menggunakan spektrogram. Menggunakan MATLAB, kepekatan kimia boleh dikira berdasarkan perkarangan penyelesaian. Dengan menggunakan asid akrilik sebagai contoh, yang mana asid tak tepu yang paling mudah karboksilik yang mempunyai ikatan kembar dan kumpulan carboxyl dalam C3 satu molekul dengan formula $\text{CH}_2=\text{CHCOOH}$, boleh ditentukan peratusan kepekatan sebatian adalah asid akrilik. Berdasarkan graf yang di plotkan menggunakan perisian MATLAB, luas dibawah graf dikira. Menggunakan luas kawasan tersebut, kepekatan boleh dikira secara langsung berdasarkan data daripada HPLC-kromatografi pretasi tinggi cecair.

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CHAPTER 1

INTRODUCTION

1.1 Introduction

Nowadays, there are so many chemical compounds that can be harm to us such as Chlorofluorocarbon (CFC) if the amount of it's become higher. But, there are certain compounds useful in production fields. The compound are combined with others type of chemical elements. It's also has their own concentration which we can calculate by using formula based on theory values. Concentration is defined as the abundance of constituent divided by the total volume of mixture. By the way to calculate the concentration, the method used before need a long time for verified the values that we want to have in the solutions. There are some devices that can help verified this unknown chemical solution. Based on the result from those devices, the data calculated by substitutes the values in that standard formula. In others sides, There are others method that can be used for calculated the concentration of the chemical compounds using spectrogram.

1.1.1 Spectrogram

Spectrogram is a time-varying spectral representation (forming an image). There are many devices that can produce these images in different types of graphs. The most common format is a graph with two geometric dimensions: the horizontal axis represents time, the vertical axis is frequency; a third dimension indicating the amplitude of a particular frequency at a particular time is represented by the intensity or colour of each point in the image. The devices which can form these types of signals are HPLC- High Performance Liquid Chromatography, UV-VIS (spectrophotometer), mass spectrometry and FTIR- Fourier Transform Infrared Spectroscopy. In this project, data from HPLC- High Performance Liquid Chromatography has been used to calculate the concentration of the solution.

1.1.2 Image Processing

Image processing is including image segmentation and it is crucially significant for the successfulness recognition of the image, it is still a dream for the computer to outperform human natural ability for visual interpretation, and thus feature extraction still remains a challenging task in various realms of computer vision and image analysis. There an image segmentation based on color texture measurements, texture is one of most popular features for image classification and retrieval. For as much as grayscale textures provide enough information to solve many tasks, the color information was not utilized. But in the recent years, many researchers have begun to take color information into consideration. Color Texture analysis plays an increasingly important role in computer vision. Since the color textural properties of images appear to carry useful information for discrimination purposes, it is important to develop significant features of color textures.

1.1.3 HPLC-high performance liquid chromatography

HPLC is a separation technique that involves the placement (injection of a small volume of liquid sample into a tube packed with porous particles (stationary phase), where individual components of the sample are transported along the packed tube column by liquid moved by gravity. HPLC typically utilizes different types of stationary phases (i.e. sorbents) contained in columns, a pump that moves the mobile phase and sample components through the column, and a detector capable of providing characteristic retention times for the sample components and area counts reflecting the amount of each analyte passing through the detector. Analyte retention time varies depending on the strength of its interactions with the stationary phase, the composition and flow rate of mobile phase used, and on the column dimensions. HPLC is a form of liquid chromatography that utilizes small size columns (typically 250 mm or shorter and 4.6 mm i.d. or smaller; packed with smaller particles), and higher mobile phase pressures compared to ordinary liquid chromatography.

With HPLC, a pump (rather than gravity) provides the higher pressure required to move the mobile phase and sample components through the densely packed column. The increased density arises from the use of smaller sorbent particles. Such particles are capable of providing better separation on columns of shorter length when compared to ordinary column chromatography.

1.2 Problem Statements

In this project, the solution are called “unknown solution” has been used to determined the concentration. The types of solution are acrylic acid and formic acid solutions. Its concentration determined based on the result displayed on HPLC- High Performance Liquid Chromotography screen data. Then, by using the result, that will be identified using MATLAB software by observe the image to calculate the concentration of the solution. This method provided the faster ways to calculate the concentrations of the solution.

1.3 Project Objectives

The objectives of this project are:

- I. To understand how the devices of HPLC can gives the result of spectrogram types.
- II. To applied the knowledge of MATLAB in creating coding. So, can determine the concentration based on the images from the data of HPLC.
- III. To calculate the chemical concentration based on spectrogram.

1.4 Scope of Project

To achieve the project objectives, some analysis are done. There is a study on how the HPLC-High Performance Liquid Chromatography, works and how to get the pure result of graph since there are a specified software that can only read the file of the data. This research done by using hardcopy of the result from HPLC data which can be used to plot others graph that can read by MATLAB software. Then, the structure of a compound can determine by observing its fragmentation especially on the bonding of the chemical compounds. Next, for this project, the types of solution are acrylic acid and formic acid which can be used using HPLC. This solution doesn't a pure acid since it need to go through dilute stage by dilute solution which decide before the experiment is done.

- a) $\text{CH}_2=\text{CHCOOH}$ - acrylic acid formula
- b) CH_2O_2 – formic acid formula

1.5 Expected Result

The expected results from this project are to obtain the concentration of the solution based on the area of the plotted data from HPLC using MATLAB code. Besides that, the calculation doesn't need to take long time since the data from HPLC can copy and paste to excel file. Then, the data read by MATLAB code and start calculated based on the formula given. The values are compared with calculated values by manual and theory concentrations. Mostly, the concentration is low since this solution is for students experiment and it's a part of safety.

CHAPTER 2

LITERATURE REVIEWS

2.1 Concentration

In chemistry, concentration is defined as the abundance of a constituent divided by the total volume of a mixture. Four types can be distinguished: mass concentration, molar concentration, number concentration, and volume concentration. The term concentration can be applied to any kind of chemical mixture, but most frequently it refers to solutes in homogeneous solutions [1]. A negative concentration value indicates that the sample was analyzed but that the concentration was below the determination limits of the analytical method. All instrumental methods for analyzing the concentrations of various elements in sediments, waters, rocks, or plants have lower determination limits. Concentrations below this lower determination limit are too small to be detected by the method. For example, one of the Oak Ridge Multielement Emission Spectrochemical Analysis methods could not detect concentrations of silver (Ag) that were less than 2 parts-

per-million (ppm). When the analytical method determined that a sample contained less than 2 ppm Ag, a value of "-2" was recorded in the Ag_ppm field. Unfortunately, we don't know if that means that the sample contains 1.8 ppm Ag or 0.0001 ppm Ag. The best that can be said is that the concentration is <2 ppm Ag. Some analytical methods record different negative numbers for an element in successive samples [2].

2.2 Spectrogram

Spectrogram is a time-varying spectral representation (forming an image) that shows how the spectral density of a signal varies with time [3]. There are many devices that can produce these images in different types of graphs. The devices which can form these types of signals are HPLC- High Performance Liquid Chromatography, UV-VIS(spectrophotometer), mass spectrometry and FTIR-Fourier transform infrared spectroscopy.

2.3 High Performance Liquid Chromatography-HPLC

HPLC is basically a highly improved form of column chromatography. Instead of a solvent being allowed to drip through a column under gravity, it is forced through under high pressures of up to 400 atmospheres. That makes it much

faster. It also allows you to use a very much smaller particle size for the column packing material which gives a much greater surface area for interactions between the stationary phase and the molecules flowing past it. This allows a much better separation of the components of the mixture [4]. *The principles of Chromatography* is a technique by which a mixture sample is separated into components. Although originally intended to separate and recover (isolate and purify) the components of a sample, today, complete chromatography systems are often used to both separate and quantify sample components. The term, "chromatography" was coined by the Russian botanist, Tswett, who demonstrated that, when a plant extract was carried by petroleum ether through a column consisting of a glass tube packed with calcium carbonate powder, a number of dyes were separated, as shown in Figure 1. He named this analysis method "Chromatographie" after "chroma" and "graphos", which are Greek words meaning "color" and "to draw," respectively[5].

2.4 UV/Vis Spectroscopy

UV/Vis spectroscopy is routinely used in analytical chemistry for quantitative determination of different analytic, such as ion, compounds, and biological macromolecules. Spectroscopic analysis is commonly carried out in solutions but solids and gases may also be studied [6]. Solutions of transition metal ions can be colored (i.e., absorb visible light) because d electrons within the metal atoms can be excited from one electronic state to another. The color of metal ion solutions is strongly affected by the presence of other species, such as certain anions [6]. Mass

spectrometry (MS) is an analytical technique that measures the mass-to-charge ratio of charged particles. It is used for determining masses of particles, for determining the elemental composition of a sample or molecule, and for elucidating the chemical structures of molecules, such as peptides and other chemical compounds [7]. The combination of a mass spectrometer and a gas chromatograph makes a powerful tool for the detection of trace quantities of contaminants or toxins. A number of satellites and spacecraft have mass spectrometers for the identification of the small numbers of particles intercepted in space. Mass spectrometers are used for the analysis of residual gases in high vacuum systems. An atom probe is an instrument that combines time-of-flight mass spectrometry and field ion microscopy to map the location of individual atoms [8].

2.5 Fourier transforms infrared spectroscopy (FTIR)

Fourier transform infrared spectroscopy (FTIR) is a technique which is used to obtain an infrared spectrum of absorption, emission, photoconductivity of a solid, liquid or gas. An FTIR spectrometer simultaneously collects spectral data in a wide spectral range. This confers a significant advantage over a spectrometer which measures intensity over a narrow range of wavelengths at a time [9].

2.6 Image Processing

An image processing is any form of signal processing for which the input is an image, such as a photograph or video frame. The output of image processing may be either an image or a set of characteristics or parameters related to the image. Most image processing techniques involves treating the image as two dimensional signals and applying standard signal processing techniques to it. Recently, a lot of works was done by depending on the computer, in order to let the processing time to be reduced and to provide more results that are accurate, for example depending on different types of data such as digital image and characters and digits. In order to automate systems that deal with numbers such as fingerprint verification, face recognition, iris discrimination, chromosome shape discrimination, optical character recognition, texture discrimination, and speech recognition. Digital image recognition has been extremely found and studied. Various approaches in image processing and pattern recognition have been developed by scientists and engineers to solve this problem (Al Omari, 2009)[10].

CHAPTER 3

METHODOLOGY

3.0 Introduction

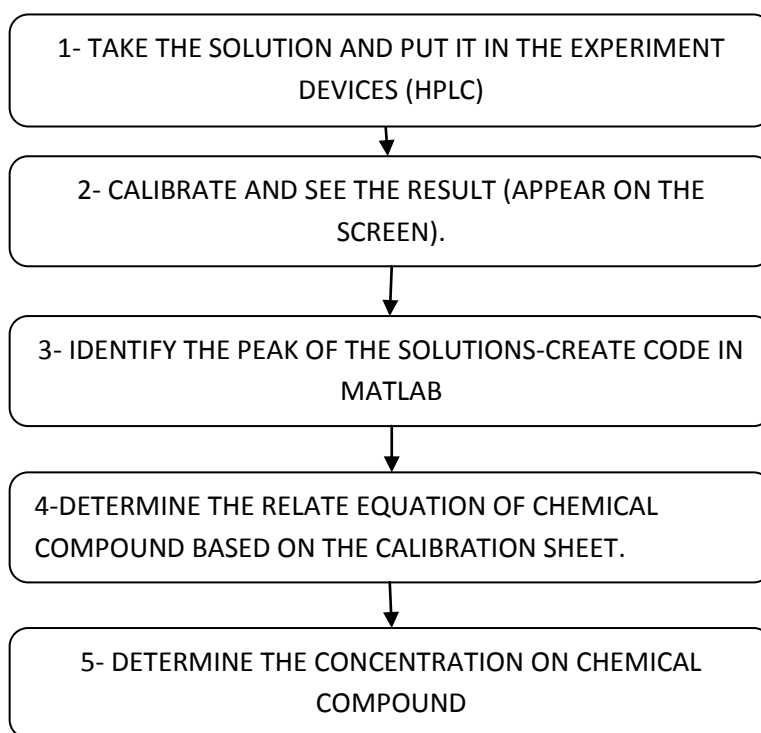


FIGURE 3.1: Flow chart for method and instructions use in this project

Based on the flow chart above, the first step of calculate the concentration by using spectrogram are by taking the unknown solution which we does not know the concentration of chemical used. Then, put it in the experiment equipments (HPLC-High Performance Liquid Chromatography). Adjust the devices for not given any error when the measurements are done. Next, calibrate the device and the results of the signal appear on the screen. Then, see the peak of signal for determined what kind of chemical elements exist in that solution. Based on the signal shown, we can identify concentration of every compound in solution used. That because, different compound give different values of concentration. By referring to these values, we can determine the compound. Then, after the results of the spectrum of every compound got, we use those results for determined the concentration of that compounds. Lastly, we also can identify the chemical elements existing in the solution we use. Continuing with step before, by using the signal and MATLAB software, create a code to calculates the concentration using the calibration theory based on the area of the peak signal. For getting the signal, it needs to plot graph using the data from HPLC and the area of the graph calculated. Next, using the area of the graph, the concentration calculated by substitute the values directly into formula. Lastly, the concentration of the solution determined.

3.1 data collection

At this time of measurement are using acrylic acid, $C_3H_4O_2$ and formic acid, CH_2O_2 as the solution of the research. For acrylic acid, there are 10 different of unknown concentration are used. Then, for formic acid, there are 5 different of unknown concentration used. Both solutions indicate using HPLC.



Figure 3.2: Data collect from HPLC- High Performance Liquid Chromatography

By using the HPLC, the data collect and the result analyze based on the graph shape and determine whether the data plotted are same or not with the values tables. Since the device have technical problems to produce the detail, in this project just used the hardcopy of results by others student before this.

3.2 plotting Graph

Since there is a problem to generate softcopy of the image from HPLC, a new graph need to plot based on the data given. Even the graph plotted doesn't exactly same with the original, but the graph plotted truly based on the data directly. The

data write in excel file. There was no graph paper or manual calculation, just using technology application which is can be done by using MATLAB software code.

For plot a graph (figure 3):

Code:

```
y= xlsread('data1.xlsx','y=mAU')
x= xlsread('data1.xlsx','x=time (m) ')

plot(x,y)
grid minor
```

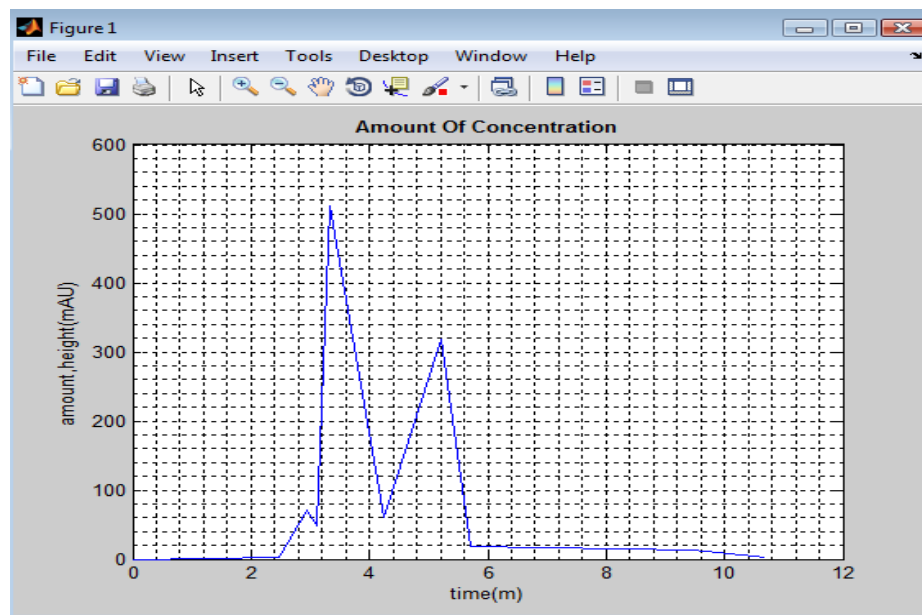


Figure 3.3- new graph plotted by using data

3.3 Area

Next step, using other code of MATLAB, determined the area under the graph. For area under graph, code **trapz(x,y)** are used. It's doesn't means the graph shape are trapezoid. **Trapz(x,y)** code is computes an approximation of the integral of Y via the trapezoidal method (with unit spacing). To compute the integral for spacing other than one, multiply Z by the spacing increment. Input Y can be complex.

The codes are:

```
AUC=trapz(x,y)

title('Amount Of Concentration','fontweight','bold')
xlabel('time (m)'); ylabel('amount,height (mAU)')
```

3.4 Concentration

Basically, from theoretical values of concentration of acrylic acid, $C_3H_4O_2$ is between range 10-0.1 mg/l and formic acid, CH_2O_2 is 8.518 g/l. By using the code from MATLAB, the concentration calculated are between range and lower. Although the concentration are lower, but, the hazardous still exist to human expecially to whow who not pay attention to safety when handling this types of solution.

The code is:

```
m=13.03438      %y=mx+c
c=139.94412     % x(concentration)=AUC-c/m

z=(AUC-c)/m     %concentration of arcylic acid
                %unit for concentration= ng/ul, n=10^-9

m=13.03438      %y=mx+c
c=139.94412     % x(concentration)=AUC-c/m

z=(AUC-c)/m     %concentration of formic acid
                %unit for concentration= ng/ul, n=10^-9
```