Design and development of low cost Nano drop UV-Vis spectrometer

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Abstract. The ultimate aim of the project is to develop a low-cost spectrometer that analyses the samples in the Nanoscale range and to minimise the usage of samples for diagnostic application. The reflection, absorption or transmission phenomenon alters the incident light during the interaction with the sample. A spectrometer measures this change over a range of incident wavelengths of electromagnetic radiation that has interacted with a sample. The light that acts as a source is passed through the sample. The prepared test sample is analysed in order to validate the developed system. Following the test samples, urine is analysed. 10 volunteers were involved as subjects. The light from the sample passes through a slit and then reflected by a collimating mirror. The wavelength of the light from the sample is analysed by Thermino software in the UV and visible range. The input to this software is provided by the webcam. A 1000 lines/mm diffraction grating is utilized to split the light into its constituent wavelength. This is a low-cost system than the available commercial spectrometers used in the laboratory.

Key words: Spectrometer, UV-Vis, Electromagnetic Spectrum, Urine Analysis, Wavelength.

1. General introduction
A spectrometer analyses the spectrum of a substance. The ultimate aim of a spectrometer is to quantify the interaction or emission of electromagnetic radiation (EM) with or from the sample. The interaction of EM radiation gives rise to three phenomenon such as absorption, reflection and scattering. The emission that occurs when the energized atoms in the samples releases the gained energy in the form of fluorescence, phosphorescence, electroluminescence. The spectrometer developed is associated to examine the UV and visible light of electromagnetic radiation [4][5].

| Energy (eV)       | 10⁸  | 10⁶  | 10⁴  | 1   | 10²  | 10⁴  | 10⁶  | 10⁸  | 10⁸ |
|-------------------|------|------|------|-----|------|------|------|------|------|
| Gamma rays        |      |      |      |     |      |      |      |      |      |
| X-rays            |      |      |      |     |      |      |      |      |      |
| UV rays           |      |      |      |     |      |      |      |      |      |
| Visible light     |      |      |      |     |      |      |      |      |      |
| IR rays           |      |      |      |     |      |      |      |      |      |
| Radar             |      |      |      |     |      |      |      |      |      |
| FM                |      |      |      |     |      |      |      |      |      |
| TV                |      |      |      |     |      |      |      |      |      |
| Short Wave        |      |      |      |     |      |      |      |      |      |
| AM                |      |      |      |     |      |      |      |      |      |
| Wavelength (m)    |      |      |      |     |      |      |      |      |      |

The spectrometer results are represented in terms of wavelength. The wavelength of light is critical since the atoms in the samples are specific to a particular wavelength. Optical filters are used to for
wavelength selection if a specific wavelength is to be passed through the sample [1]. Diffraction grating is a dispersive element used to present the polychromatic light that is incident on it as a spectrum. The diffraction grating is based on the principle of constructive and destructive interference. The range of wavelength for each colour in the visible spectrum is shown in table 1 [10].

| COLOUR  | WAVELENGTH (nm) |
|---------|-----------------|
| Violet  | 380-450         |
| Blue    | 450-495         |
| Green   | 495-570         |
| Yellow  | 570-590         |
| Orange  | 590-620         |
| Red     | 620-750         |

2. Methodology
2.1 Introduction
A spectrometer is used to analyse the electromagnetic spectrum from a light source. In the developed system the light is passed through a sample and its spectral components are obtained.

The experiment is carried out inside a black paint coated acrylic chamber. The sample is placed in a pit made on the surface of the board. The light source is placed above the sample outside the chamber. The light collected after passing through the sample is sent into a spectrograph inside the chamber. The spectrograph consists of a diffraction grating and a webcam. The diffraction grating separates the light into a spectrum consisting of several wavelengths and a webcam records the data. A spectrometer uses a mirror to produces a collimated beam of light before it is being dispersed by grating. The spectrum that falls on the webcam is simultaneously measured. The recorded data is analysed by the Theremino software.

Ten normal male volunteers of 21-25 years old were studied. Their mean Body Mass Index (BMI) was 23.06±2.45 kg/m². The urine was collected in a sterile container. The individuals were instructed to collect the urine before breakfast as the intake of food may cause a change in colour of urine. 2 out of 10 subjects have increased glucose level. The volunteers do not have any history of liver failure, bladder infection, metabolic disorder and kidney infection.

![Figure 2. Block Diagram of the Developed System](image)
The components of the developed spectrometer are a light source, a pit for holding sample, a diffraction grating and a webcam as detector.

2.2 Source
The LED as a light source is encouraged [6]. The white LED comprises of all the wavelength suitable for the UV-Vis spectrometer [2]. The white LED has improved luminous efficacy, longer life span, high colour rendering index, correlated temperature values.

2.3 Sample preparation
The urine is analysed using the spectrometer developed. Initially, the test samples are analysed so as to validate the system. The test sample is prepared by mixing 1 gram of the selected crayon in 5ml of water. This is opted to have a concentrated solution. The experiment is carried out inside an acrylic board chamber of 3mm thickness. It is less expensive and more durable. The acrylic board is made into desired dimension with laser cutting for the purpose of assembling them. A concave depression is made on the superior surface of the board to place the sample. The sample used in the biochemistry lab is wasted for the diagnosis. In an effort to minimize the quantity of the sample used, the process should be carried out in Nanoscale range. The 500 nanoliter out of the prepared 5ml sample is placed on the concave surface with the help of a digital micropipette. The digital micropipette of 0.5 -12.5 µl range is used. 500 nl is drawn by adjusting volume indicator by rotating the plunger. The light after passing through the sample enters a slit placed inside the acrylic chamber. The amount of light that enters the closed chamber is determined by the slit. The slit is followed by a collimating mirror. The diverged light is collimated by the concave surface of the mirror.

2.4 Spectrograph
The diffraction grating is the principal optical element in the developed system. The light that consists of all wavelength is separated into its component wavelengths by diffracting them at various angles [2]. The diffraction grating used in this project is of 1000 lines/mm [7]. The component wavelength ranges between 300-700 nm. This comprises a part of UV and complete visible spectrum. The webcam used is a Logitech C270 HD Webcam [8]. This is a PC webcam with a connectivity of USB. Thermino spectrometer is an open software that offers the user to look the constituent wavelength of light that emerges out from the diffraction grating. The GUI provides the information of the wavelength in real time. It allows to choose the USB [9].

3. Results and discussion
The test sample analysed has a wavelength of the electromagnetic spectrum in the visible range. The wavelength produced by the test sample is given in the figure 3. The test samples are prepared and analysed for spectrum in the effort of validating the developed spectrometer. Followed by the test sample, urine collected from the volunteers were analysed. Figure 4 shows the wavelength of the urine collected from each subject. Since the patients have no history of liver failure, kidney and bladder infection the colour of urine ranges from very pale yellow to pure yellow. The average of the wavelength obtained from the urine sample is 574.7±3.65 nm.

In biomedical application the sample are wasted in the testing and diagnosing process. In order to minimise the samples used, it's appropriate to test with sample in Nano scale. Parallel operations are possible with their Nano drop and many number of trials can be carried out for the samples.

For calibration, the spectrometer is tested with a colour LED. The wavelength of the colour LED should be known. Since the wavelength is analysed with Thermino, auto-correction is possible with the software.

The urine is chosen as sample because a further analysis is to be carried out to diagnose Polycystic Ovarian Syndrome (PCOS) at low cost and in real-time. The wavelength obtained from the urine using spectrometer is to be considered as a major parameter to detect PCOS.
4. Conclusion
Spectroscopic analysis is carried out in solutions especially for biological fluid in nanoscale range. This makes it possible to reduce the amount of sample used in biochemistry lab. The development of this spectrometer has the potential to replace the commercially available spectrometer. This device is developed to increase the efficiency of biological discovery. An integrated, low cost system capable of performing complex biological protocol can be realised in real time. The urine is selected as sample since it is further to be used for Polycystic Ovarian Syndrome (PCOS) diagnostic in another project.

5. References
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