

Quantitative Analysis of C - Reactive Protein (CRP) Using Quantum DOTs

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Abstract: - In this study, a method of analyzing blood serum and quantification of C-reactive protein (CRP) using quantum dots is proposed. Blood serum is the one that contains many protein including CRP, and hence analyzing this could be helpful in diagnosing any disorder at the earliest. In this study, two experiments are done; one test on blood serum and the other on CRP. Initially, blood serum is separated from the blood and is taken in test tube is successively diluted using NaCl solution. These samples are illuminated using standard white light and the transmitted light is picked up from the color sensor (TCS2300). Similar kind of test is also performed on CRP samples at different concentration level. RGB values obtained from these two tests changes as the concentration of blood serum and CRP changes. Quantification of these colors would represent CRP concentration or concentration of protein content in the blood. Hence, by precisely calibrating these color changes against protein concentration would result in a device that could quantify blood chemical parameters.

Index Terms— Blood serum, Color sensor, Quantum dots (CdTe) , Lighting, LED

I. INTRODUCTION

C-reactive protein (CRP) is an annular (ring-shaped), pentameric protein found in blood plasma, whose levels rise in response to inflammation. CRP was so named because it was first identified as a substance in the serum of patients with acute inflammation that reacted with the C-polysaccharide of Pneumococcal. It is a blood test that measures the amount of a protein called C-reactive protein in our blood. C-reactive protein measures general levels of inflammation in our body. High levels of CRP are caused by infections and many long-term diseases. But a CRP test cannot show where the inflammation is located or what is causing it. Other tests are needed to find the cause and location of the inflammation. CRP is synthesized by the liver in response to factors released by macrophages and fat cells (adipocytes). It is a member of the pentraxin family of proteins. It is not related to C-peptide (insulin) or protein C (blood coagulation). Under normal conditions, the baseline concentration of CRP in the plasma is around 0.8 mg/L. There is a high risk of developing cardiovascular disease if CRP level is higher than 3 mg/L, average risk if in the range 1–3 mg/L and less risk if less than 1 mg/L. CRP is a member of the class of acute-phase reactants, as its levels rise rapidly during inflammatory processes occurring in the body. During the acute-phase response, levels of CRP increase rapidly reaching a peak at 48 hours.

When the inflammatory processes stop, levels of CRP decline with a relatively short half-life of 18 hours. There are a number of methods for detecting CRP in blood such as immunonephelometry method electrochemical technique using gold electrode surface, antibody-conjugated magnetic nanoparticles enzyme-linked immunoabsorbent assay (ELISA), turbidimetric assay, chemiluminescent detection based on gold nanoparticles.

Quantum dots (QDs) are very small crystals of semiconductor materials. Their size ranges from about a hundred to a few thousand atoms. The diameter of a quantum dot is approximately between two and ten nanometers, which puts them in a special size range that retains some properties of bulk materials, as well as some properties of individual atoms and molecules. Quantum dots are semiconductor nanocrystals that have tunable emission through changes in their size. Producing bright, efficient quantum dots with stable fluorescence is important for using them in applications in lighting, photo voltaics, and biological imaging. This study aimed to optimize the detection of CRP in blood serum using a NaCl with distilled water and set of test QDs(CdTe) .The test has been performed with or without QDs. First, a small quantity (0.75ml) of saline , 1 ml of blood serum S1 was equally divided into different parts. All the different samples of blood serum at different concentrations levels were obtained.

Similarly, sample taken from S1 i.e (another small

quantity (1ml) of blood serum with 0.75 ml of saline) was equally divided as per the above procedure. Each of the samples was added 0.04 ml of QDs(CdTe) for successive dilution. Further, this test is analyzed by various characterization like LASER and white light with 0.75 ml of saline (NaCl), 1 ml of QDs and 0.04 ml of CRP solution with positive and negative agglutination. Each of the sample was added 0.04 ml of CRP .These QDs are studied for their size, structure, bonding as well as optical characteristics.

II. EXPERIMENTAL

a. Chemicals used

Human CRP and CRP-free human serum were obtained from Biotrend, Germany. MPC was purchased from TCI, Germany. Parylene C dimer (di(2-chloro-p-xylylene)) was supplied by Specialty Coating Systems, USA.

CRP sample concentration was half of the respective CRP serum concentration. CRP samples with inhibitor for binding inhibition assays contained 0.5 mM cytidine 5-diphosphocholine (in addition to the respective CRP concentration) and were left to incubate for 30 min prior to the experiment.

b. Instruments

The TCS230 is Programmable Color Light-to-Frequency Converter Module. It can be controlled in many ways, one of which is Arduino. Here, RGB component values are not equal. For TCS230 module the sensitivity of the three light is different, which leading to RGB value different.

c. Addition of CDSE for successive dilution

In this method , LASER of 650 nm and white light emitting diode (LED) are used as a light source . The sample (serum and saline) is placed in between the LED light source and sensor .Then the sample along with the whole set up is covered in a black body .

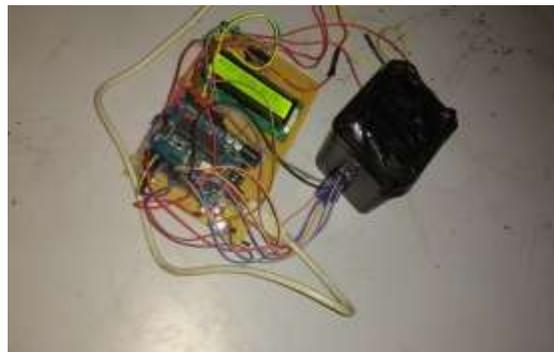


Figure 1. Electronic circuit for color sensing

The blood serum is successively diluted with saline to form samples with different concentrations.



Figure 2. Successive dilution

d. Detection of CRP

CRP could also be detected using immunoassay methods involving factors such as the availability of antibodies, their reproducibility and costs, as well as the discrepancies in results found when polyclonal versus monoclonal antibodies are used. Currently used methods for detection of CRP in clinical laboratories are mostly based on latex agglutination or immuno nephelometry. This test is analyzed by various characterization like LASER and white light with 0.75 ml of saline (NaCl), 1 ml of QDs and 0.04 ml of CRP solution with positive and negative agglutination . Each of the sample was added 0.04 ml of CRP.

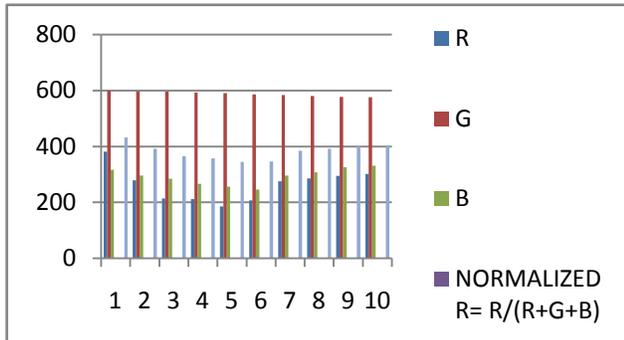


Figure 3. change in concentration with CRP using QDs.

III. RESULTS AND DISCUSSIONS

Using successive dilution technique ,known concentrations of blood serum was diluted with known concentrations of water. LASER input with the intensity of 650 nm was given and the corresponding equivalent output voltages were measured. The voltages correspond to the total amount of serum protein present in the blood.

Test which is done with or without QDs is shown in Figure 4. A small quantity (0.75 ml)of saline , 1 ml of blood serum S1 was equally divided into different parts. All the different samples of blood serum at different concentrations levels were obtained. Similarly, sample taken from S1 i.e (another small quantity (1ml) of blood serum with 0.75 ml of saline) was equally divided as per the above procedure. Each of the samples was added 0.04 ml of QDs(CdTe) for successive dilution.



Figure :4 . Different concentrations of successive dilution

The CDTE QDs are analysed by various characterization like LASER and white LED.

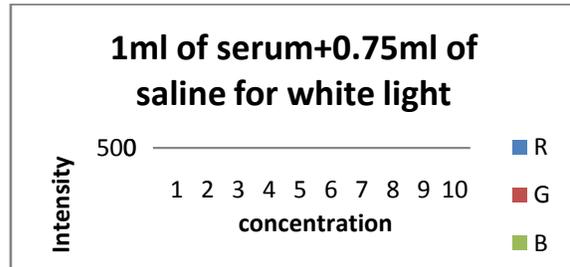


Figure 5a. change in concentration using white light without QDs.

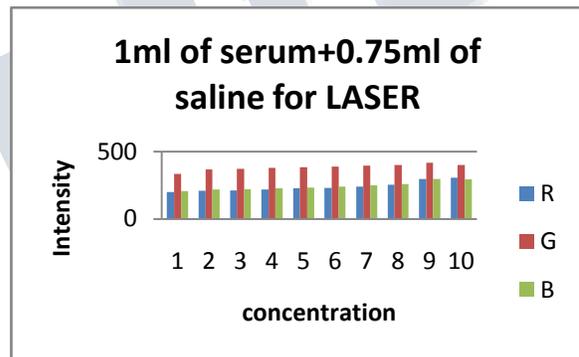


Figure 5b. change in concentration using LASER without QDs.

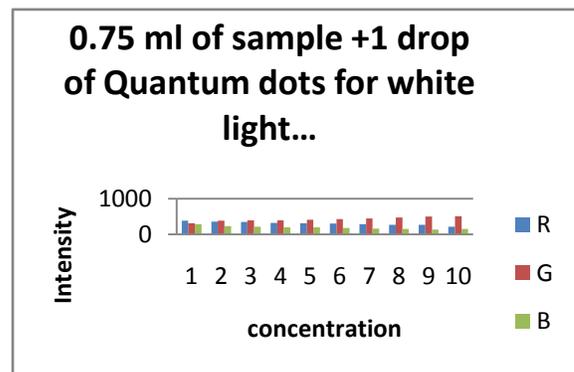


Figure 5c change in concentration using white light with QDs.

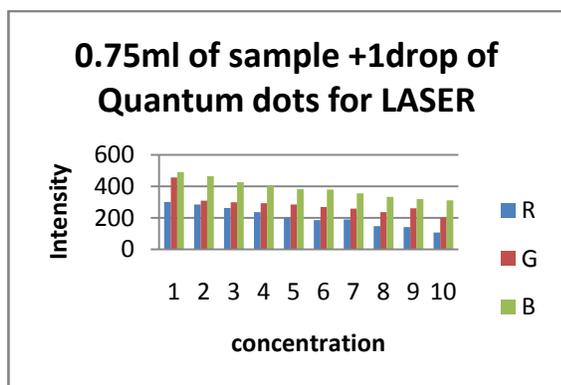


Figure 5d change in concentration using LASER with QDs.

IV. CONCLUSIONS

This study was able to optimize simple and rapid method for the detection of CRP can be developed using CdTe quantum dots. An attempt was made to determine the ratio of the CdTe shell through fluoroscopy and it was found that the method can determine the particle size.

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