SYNTHESIS AND CHARACTERISATION OF pH SENSITVE CARBON DOTS BY CARBONISATION OF GLUCOSE

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Submitted by

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CERTIFICATE

I certify that this is bonafide record of the project work performed by Nandu Krishnan M S, Devika S and Gopika Rajesh in partial fulfillment of the requirement of Bachelor Degree in Chemistry of the Kerala University.This work is carried out under my guidance and that no part of this work has been reported for any other degree.

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DECLARATION

We hereby declare that this project work entitled **“Synthesis and characterization of pH sensitive carbon dots by carbonization of glucose’’** submitted to Kerala University in partial fulfilment of Bachelor’s Degree in Chemistry is bonafide record of the work carried out under the guidance of Assistant Professor Ms. Linda E Jacob and no part of it has been submitted for any other degree or diploma.

CONTENTS

ABSTRACT

Carbon dots are new series of fluorescent nanomaterials that have drawn great attention in recent years owing to their unique properties. In this project, a simple carbonization approach to synthesize carbon dots was developed by using glucose as the carbon precursor. The synthesized carbon dots were further characterized using UV-Visible spectra. The effect of pH on the UV-Visible spectra of carbon dots were also analysed. The aqueous solution of as-prepared carbon dots were nearly transparent under visible light, while it displayed strong yellow fluorescence under UV light (365nm). Significant changes in the absorbance of the carbon dots were observed as the pH of the medium changes from acidic to basic implying that the as-prepared carbon dots were very sensitive to pH. So the carbon dots can be used as a promising material for sensing, bio imaging etc.

INTRODUCTION

NANOTECHNOLOGY

The emergence of nanotechnology as a field in the 1980s occurred through convergence of Dexler's theoretical and public work,which developed and popularized a conceptual framework for nanotechnology and high-visibility experimental advances that drew additional wide-scale attention to the prospects of atomic control of matter.

Nanotechnology (nanotech) is the manipulation of matter on an atomic, molecularand supramolecular scale. The earliest, widespread description of nanotechnology referred tothe particular technological goal of precisely manipulating atoms and molecules for fabrication of macro scale products,also now referred to as molecular nanotechnology.

Nanotechnology as defined by size is naturally broad,including fields of science as diverseas surface science,organic chemistry,molecular biology,semiconductor physics,energystorage ,engineering, micro fabrication,and molecular engineering.The associated research and applications are equally diverse,ranging from extensions of conventional device physics to completely new approaches based upon molecular self-assembly, from developing new materials with dimensions on the nanoscale to direct control of matter on the atomic scale.

The study of nanotechnology has resulted in the introduction of various types of nanomaterials, which possess diversified properties that can lead to the production of intriguing properties.Carbon nanodots, also called carbon dots are a class of carbon nanoparticles that function within a nano system.They have recently gained significant attention due to their feasible and varied synthesis methods,unique optoelectronic,and strong luminescent behavior.

CARBON DOTS

Carbon dots (CDs),a relatively new member of the carbon nanomaterial family,were firstobtained during purification of single walled carbon nanotubes in 2004 **(1)**. They are generally oxygenous carbon nanoparticles with size of less than 10nm. CDs have subsequently attractedconsiderable attention due to their simple synthesis procedure combined with fascinatingphysical properties **(2).** Just like heavy-metal-based quantum dots (QDs),they exhibit severalpromising advantages over organic fluorescence dyes,such as tunable luminescence emission,high stability against photo bleaching and blinking.In addition,CDs are biocompatible and smalldots with low molecular weight and low toxicity,which makes them superiort of metal quantum dots**(3-5).**CDs have been applied widely in fields of catalysis**(6),**printinginks**(7),**biologicalsensors**(8-10),**bioimaging**(11)**and drugdelivery**(12).**

CDs,one of the essential fluorescent carbon nanoparticles,are earning increasing consideration in everyday human life for their practical applications.Generally speaking,CDs area new category of zero- dimensional carbon based nanoparticles that can be allocated into three group saccording to their structural manner:CDs,carbon nanodots (CQDs) and grapheme quantumdots(GQDs)**.**

CDs are amorphous quasi-spherical fluorescent carbon nanoparticles with no quantum confinement effect (QCE),in which the band gaps are independent of their size;CQDs are comprised of crystalline,quasi-spherical fluorescent carbon nanomaterials with a diameter between 1nm and10nm and possess a moderate QCE,wherethebandgapsis influenced by the CQD size;GQDs are carbon nanoparticles that consist of several layers of zero- dimensional graphene sheets with sizes between 1nm and 10nm that exhibit strong QCE,where their band gaps strongly rely on their sizes**(13).**

CDs(firstly identified as ‘carbon nanoparticles’)were accidently found by Xu et al. in 2004during the separation and purification of single-walled carbon nanotubes (SWCNTs) and triggered subsequent studies in both technological and scientific fields**(14).**This invention also attracted significant attention in discovering CDs’ fluorescence behaviours,leading to developments in the field of CD research.The discovery of CDs also attracted the interest of various researches to explore the structurally related cousins of CQDs(e.g.,carbonized polymerdots(CPDs)),which are characterized based on their structural differences,formation mechanism and properties**(15).**In 2006,these newly discovered fluorescent carbon nanomaterials were titled ‘carbon quantum dots’ by Sun et al.,who successfully disclosed a synthetic track to develop CQDs with much-enhanced fluorescence emission**(16).**

SYNTHESIS

The various routes have been proposed to fabricate the CDs with desired properties for a particular application during the last decade. Generally, these well-established synthesis strategies of CDs are classified into“top-down” and“bottom-up”.

The top-down strategies include ultrasonic synthesis, chemical exfoliation, electrochemical oxidation, arc-discharge, and laser ablation.The entire breakdown mechanism depends on the breakdown of the bond between the carbon atom.In the green synthesis of carbon dots,researches use some carbon waste materials such as candle soot,natural gas soot as precursors.These largesized carbon materials can be oxidized and broken into nano sized CDs bystrong oxidizing agents such as H_2SO_4 and HNO_3 limited raw materials,top-down method is less applied in green synthesis.

TOP-DOWN APPROACHES:

Chemical Exfoliation is a facile and convenient method for mass production of high quality CDs without complicated devices.The precursor carbon materials are cleaved by strong acids or oxidizing agents. Mao and coworkers fabricated for the first time fluorescent GQDs with different sizes from candle soot by using HNO_3under a relatively high temperature in2007**(17).**Afterwards, Penget al. prepared GQDs trough a chemical exfoliation of carbon fibresby$H_2SO_4$and HNO_3**(18).**The prepared GQDs with different sizes show yellow,green and blue PL emission under different stirring temperatures indicating that the successful preparation of GQDs can be achieved by chemical cleavage of carbon fibre.

Laser ablation, as a unique and promising synthesis route, has been applied in the preparation of CDs owning to advantages of short period and simple operations. Sun et al.first demonstrated laser ablation synthesis of GQDs from graphite**(19).**Li et al. prepared GQDs with visible,stable and tunable PL performance by laser rapid passivation of carbon particles,and demonstrated that passivation by laser irradiation has an important influence on the origin of PL**(20).**

Ultrasonic-assisted treatment is acknowledged that the method has the advantages of low cost and simple operation for the preparation of CDs.Alternate high-pressure waves and low-pressure were generated in the ultrasound process,which results in the formation and collapse of small bubbles in liquid.Thus macroscopic carbon materials were cut into nanoscale CDs by strong hydrodynamic shear forces.Generally,researches prepared the CDs with different properties by simply adjusting the ultrasonic power,reaction time and the ratio of carbon sources and solvents.

BOTTOM-UP APPROACHES

Microwave Synthesis, a green, cost-effective strategy, is widely employed to synthesize the CDs in less time.Microwave irradiation can provide uniform heat for formation of CDs.Lietal .synthesized green fluorescent GQDs by microwave-assisted chemical cleavage of GO sheets under acidic conditions for the first time **(21).** Wang and coworkers reported a simple one-pot microwave-assisted approach to fabricate the water-soluble CDs from protein-rich egg shell membranes**(22).**

Hydrothermal strategy for the preparation of CDs has advantages of low cost and nontoxicity.Compared to other synthetic routes, hydrothermal method is a simple approach to synthesize CQDs. In general, the water solution of mixtures was enclosed with Teflon in an oven and hydrothermally reacted at high pressure and high temperature.Pan et.al.reported a green route to prepare blue fluorescence CQDs by the hydrothermal method for the first time **(23).**

Solvothermal approach for the preparation of CDs has advantages of low-cost and requirement of simple equipment.Different from the hydrothermal method,water solution was replaced with one or several solvents sealed with Teflon equipped with a steel autoclave.

Fan et al. first proposed **chemical vapor deposition method** to prepare CQDs by using methane as a carbon source**(24).**In a CVD technique,the size of the ultimate product could be determined by tuning these parameters including carbon source, growth time, flow rate of the hydrogen and temperature of the substrate.

APPLICATIONS OF CARBON DOTS

OPTRONICS

CQDs possess the potential in serving as materials for dye synthesized solar cells,organic solar cells,supercapacitor and light emitting devices.CQDs can be used as photosensitizers in dye-synthesized solar cells and the photoelectric conversion efficiency is significantly enhanced.CQDs incorporated hybrid silica based sol can be used as transparent fluorescent paint.

BIOIMAGING

Bio imaging technology requires the assistance of technology, including X-ray, ultrasound and magnetic resonance imaging, to process images of living organism.Due to low/non-toxicity of CDs, they have also been considered as biocompatible fluorescent dyes for in vivo imaging instead of carrier of drug molecules. The CDs derived from different carbon sources do not abided photobleaching and photodegradation.CDs can be modified with various functional groups for appropriate PL emission. In the past years, a number of studies have reported the bioimaging potentials of CDs **(25).** The PL emission of CDs was commonly tuned to a longer wavelength range for improving signal to noise ratio (SNR).Sun et al. first proposed a route to stain Caco-2 cells by using the PEG1500N passivated CDs for cellular imaging,suggesting CDs can be utilized in fluorescent labels of cell **(26).** Subsequently, some other cells such as HeLa cells, HepG2 cells, MCF-7 cells, pancreas progenitor cells and human lung cancer cells were also used in intracellular imaging. Wei et al. prepared N-S-BCDs from allium fistulosum for cellular multicolor imaging owing to their low cytotoxicity. The surface of CDs was commonly modified with appropriate functional groups for improving absorptivity with small molecules or proteins (27). For example, CDs passivated with a hyper branched polymer showed stronger PL emission than that coated with a linear polymer.

PHOTOCATALYSIS

One of the most predominant and exciting topics in nanoscience and nanochemistry is nano-photo catalysis.CDs showed high photocatalytic activity as they can decompose organic dyes,2,4dichlorophenol, H_2O_2 anionic dye, and eosin yellow under light irradiation.TiO_2 is one of the most well-known photocatalysts and is commonly used to eliminate organic pollutants as well as generate H_2 by splitting of water **(28).** Nonetheless, a noteworthy disadvantage of TiO_2 photocatalytic efficiency is the impotent use of visible light as the illumination source. Hence,bandgap construction by possible adjustment of TiO2-based media can be done to enhance the execution of TiO2 catalysts. In this case, a nano composite of CQDs and TiO_2 is recommended to understand the effective usage of the full sunlight spectrum. For example, Li'steam designed a TiO2/CQDs complex system to exploit the employment of sunlight with a full spectrum**(29).**

They found that the TiO2/CQDs complex was able to deteriorate methylene blue (MB) under visible light illumination,whereas the control groups,which used only pure TiO2 without CQDs,showed no or little reduction of MB The result proved that CQDs are essential for effective photo degradation under visible light.

DRUG DELIVERY

One of the most interesting applications of CDs is nanomedicine.CDs,which are a type of fluorescent nanoparticles, show no toxicity under in vitro and in vivo experiments.In a typical experiment, Singh's team performed in vitro and in vivo toxicity studies of nitrogen-doped CQDs (NCQDs). In a typical in vitro experiment, lactate dehydrogenase(LDH) profile,cell apoptosis analysis, DNA fragmentation and growth cycle assessment was analysed by treating the HeLa cell line with NCQDs**(30).**The results showed no apparent toxicity of NCQDs against these cancer cell lines.

CHAPTER 2

REVIEW OF LITERATURE

Carbondots(CDs) are zero-dimensional carbon-based materials in the size range of a few tens of nanometers and can be doped with N, S, P, and B heteroatoms. They are chemically modifiable to enhance and render some additional functional properties. CDs possess many inherent assets such as tunable opto electronic properties with high photoluminescence and/orfluorescence properties, good biocompatibility, and tunable and post modifiable functional groups with facile preparation methods. As the precursor materials and synthesis. Methods greatly determine the applicability of CD materials in many fields. The approaches for CD synthesis, including the effects of source and doping as well as the reaction conditions such as temperature and time will be discussed. This will be followed by the application of CDs in bioimaging,cancertherapy,geand drug delivery,sensors and biosensors,catalysts,and energy.

Zhi Rao et al describes a method for the preparation of orange red emissive carbon atom with excitation/ emission peak at 520/582 nm. Trimesic acid and 4-aminoacetanilide were used in aone-pot hydrothermal preparation method to create the CDs. Hydrogen peroxide significantly reduces the CDs' luminescence.H2O2,which is produced when glucose oxidase (GOx) oxidizes glucose, quenches the fluorescence by static quenching. A fluorometric method for measuring glucose was developed based on this phenomenon. The method is selective for glucose over its analogues and was successfully applied to the determination of glucose in diluted human serum and in urine from diabetics and healthy individuals. Recoveries from spiked samples range from 98.7 to 102.5%.**(31)**

Ning Yuan et al A new stationary phase based on glucose-derived carbon dots-modified on silica (Sil-Glc-CDs) was prepared and applied in hydrophilic interaction chromatography.The column demonstrated greater separation selectivity than Inspire 5m HILIC column and glucose modified silica columns in the HILIC mode and was capable of separating several polarchemicals.**(32)**

Lei Wu et al blue-emitting chicken cartilage carbon dots (cc-CDs) were synthesized using biomass as green precursors. Using chicken cartilage obtained from food, we created a green,water-soluble,and extremely stable fluorescent cc-CD.For the determination of glucose ,H_2O_2,and Fe^{3+}, a multi-analyte approach was developed. Unlike in the past, a dual quenching explanation was put forth to describe the method's workings. It demonstrated great performance when compared to the published approach,including a wide line arrange,highsensitivity,
And selectivity.**(33)**

Moon-Jin Cho et al. A bienzyme (GOx and HRP) system was successfully used to create a ratiometric fluorescence technique for the detection of glucose in aqueous solutions or solid-state PAA films that had been crosslinked with DAPEG. The green-emitting Rhodamine 6G was inert to the enzymatic reaction of Glucose oxidase and Horseradish peroxide with glucose,leading in a Continuous fluorescence color change from blue to green as the glucose concentration grew in the blue-emitting Carbon Dots,which displayed fluorescence quenching**.(34)**

Zhanhui Wang et al Glucose (G-CDs) is carbonized hydrothermally to produce carbon dots(CDs). The G-CDs experience an ice-shaping effect as a result of the adsorption on ice crystals,which also prevents ice growth and recrystallization. The G-CDs are attractive cryoprotectant materials due to their excellent suppression of ice recrystallization activity and low cytotoxicity.Thus, the recovery of sheep red blood cells to about 60% can be significantly increased by GCDs without any organic solvent. These G-CDs have low cost, biocompatibility, commercial availability, and mass-production properties thanks to the utilization of glucose as a starting material, opening the door for the synthesis of both cryopreservation material CDs from sugars**.(35)**

Longyi Chen et al The fluorescence of CDs can be quenched through covalently interacting with the graphene oxide. The restored fluorescence is observed in the presence of glucose. A microwave-assisted polyol technique is created to make CDs with controllable photoluminescence in a broad emission range between 430 and 548 nm. In contrast to prior studies on excitation-dependent CDs that have been published,our method can create tunable luminescence CDs under a certain excitation by merely modifying the precursors' ratio. In contrast to our procedure,which is simple and environmentally safe,alternative techniques for making CDs frequently employ a variety of oxidative and reductive chemicals.**(36)**

MN Egorova et al Carbon precursors such as glucose and birch bark soot were used to create carbon dots by the hydrothermal synthesis process in aqueous ammonia. According to the distribution of lateral sizes of carbon dots, the average size of glucose is 10–12 nm while soot is20–22 nm. The surfaces of the produced carbon dots have oxygen groups, according to infrared absorption spectra. Aqueous solutions containing carbon dots made of glucose show high absorption in the visible spectrum between 300 and 500 nm. Carbon dots made of soot exhibit significant UV absorption but are clear in the visible spectrum. The luminescence spectra show that carbon dots made from glucose and soot are luminous in the same spectral region, that their radiation wavelengths rely on the wavelengths of excitation,and that their emission wavelengths depend on the wavelengths of excitation.Carbon dots synthesized from glucose and soot have great prospects in terms of their application in biology and medicine.**(37)**

CHAPTER 3

OBJECTIVES

1. Synthesis of carbon dots from glucose.

2. Characterization of synthesized carbon dot using UV-Vis spectroscopy.

3. To study the effect of pH on the as prepared carbon dots.

CHAPTER 4

MATERIALS AND METHODS

Materials

Chemicals

used

- □ Glucose
- □ Dilute H_2SO_4
- □ NaOH
- □ Distilled water

Apparatus and

instruments used

- □ Beaker
- □ Weighing bottle
- □ UV Lamp
- □ Glass rod

Experimental methods

1. Synthesis of carbon dots from glcose

Carbon dots were prepared by strongly heating 4g of glucose for about 15 mts in a 100ml beaker. After cooled down to room temperature, the dark brown products were mechanically ground to fine powders. After that, obtained powders were dispersed in water, and the carbon dots were collected by filtrating for further characterization and use.

Fig : carbon dots prepared from glucose

2. Characterization of the as-synthesized carbon dots

UV-Vis Analysis:

The absorption maxima of glucose carbon dotswere monitored by measuring the UV–Vis spectrum of the reaction mixture by using a UV–Vis spectrophotometer (Shimadzu model) in the wavelength ranging from 200-700 nm.

3. To study the effect of pH on the as-prepared carbon dots

The as-prepared carbon dots aqueous solutions were adjusted to the various target pH values by adding dil.H_2SO_4 or NaOH solution

CHAPTER 5

RESULTS AND DISCUSSION

In the present work, a simple carbonization approach to synthesize carbon dots was developed by using glucose as the carbon precursor. The synthesized carbon dots were further characterized using UV-Vis spectra.The effect of pH on the UV-Vis spectra of carbon dots were also analysed

Fig: Solution of carbon dots under UV lamp

The diluted solution of as prepared carbon dots in aqueous solution is nearly colorless under visible light. The photograph of the dispersion under UV light (365nm) exhibits a yellow color further revealing that the resultant dots exhibit yellow fluorescence

UV-Visible Analysis

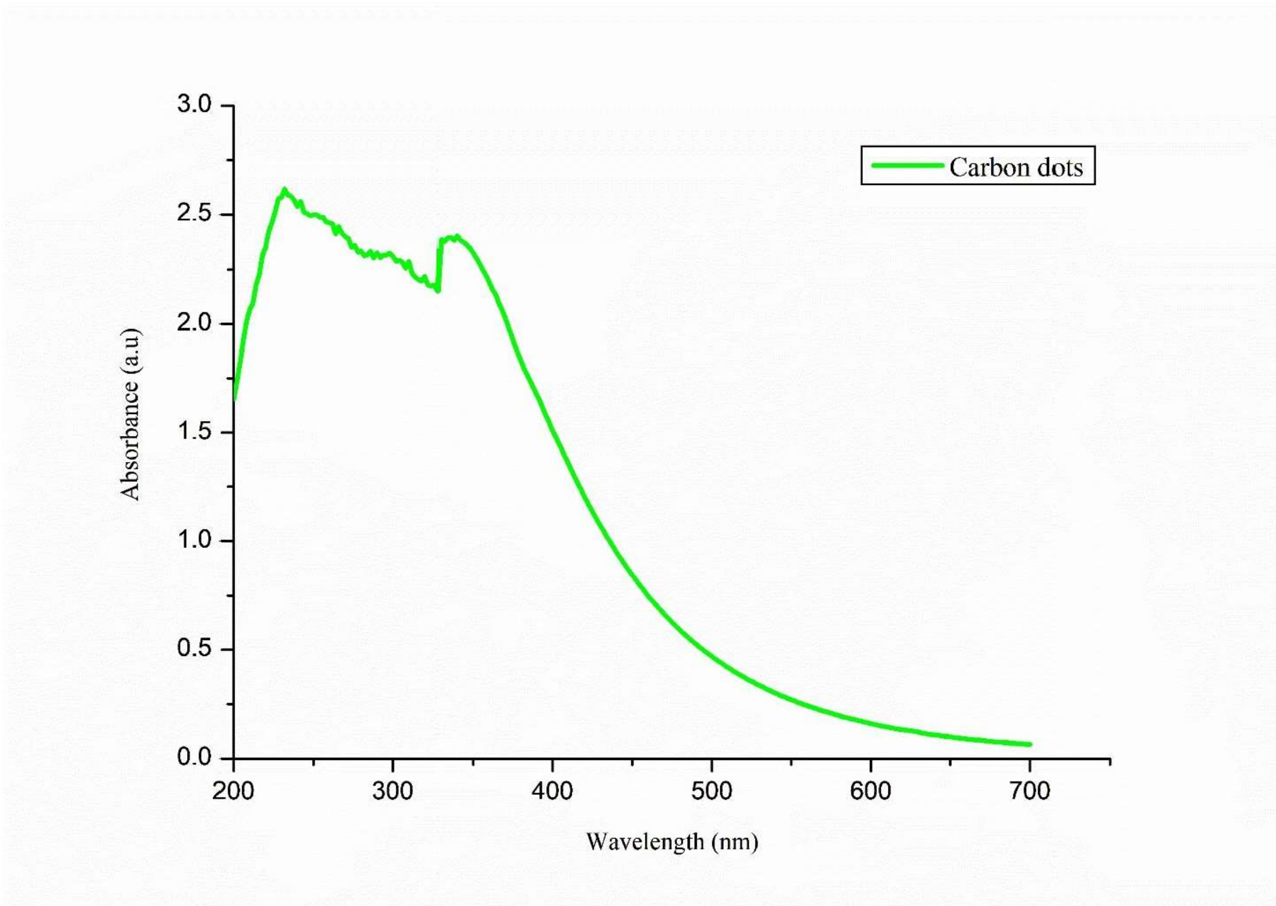

Fig: UV-Visible spectra of carbon dots

The UV- Visible spectrum of the carbon dots shows two strong peaks at 232nm and 340nm. The aqueous solution of as-prepared carbon dots were nearly transparent under visible light, while it displayed yellow fluorescence under UV light (365nm)

Effect of pH on the PL of as prepared carbon dots

Fig: carbon dots in NaOH solution (left) and HCl solution (right)

Fig: carbon dots in NaOH solution (left) and HCl solution (right) under UV lamp

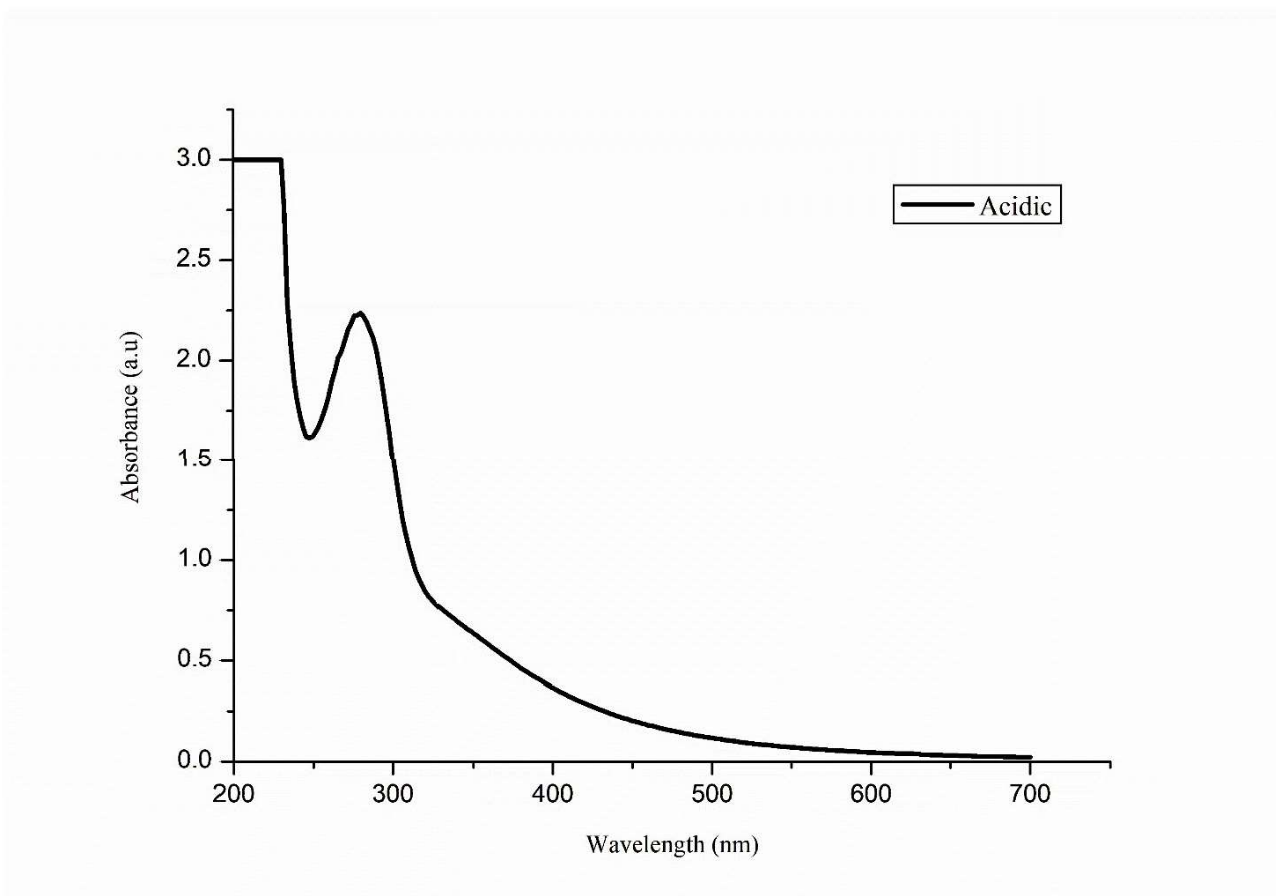

Fig: UV-Vis spectra of carbon dots in acidic medium

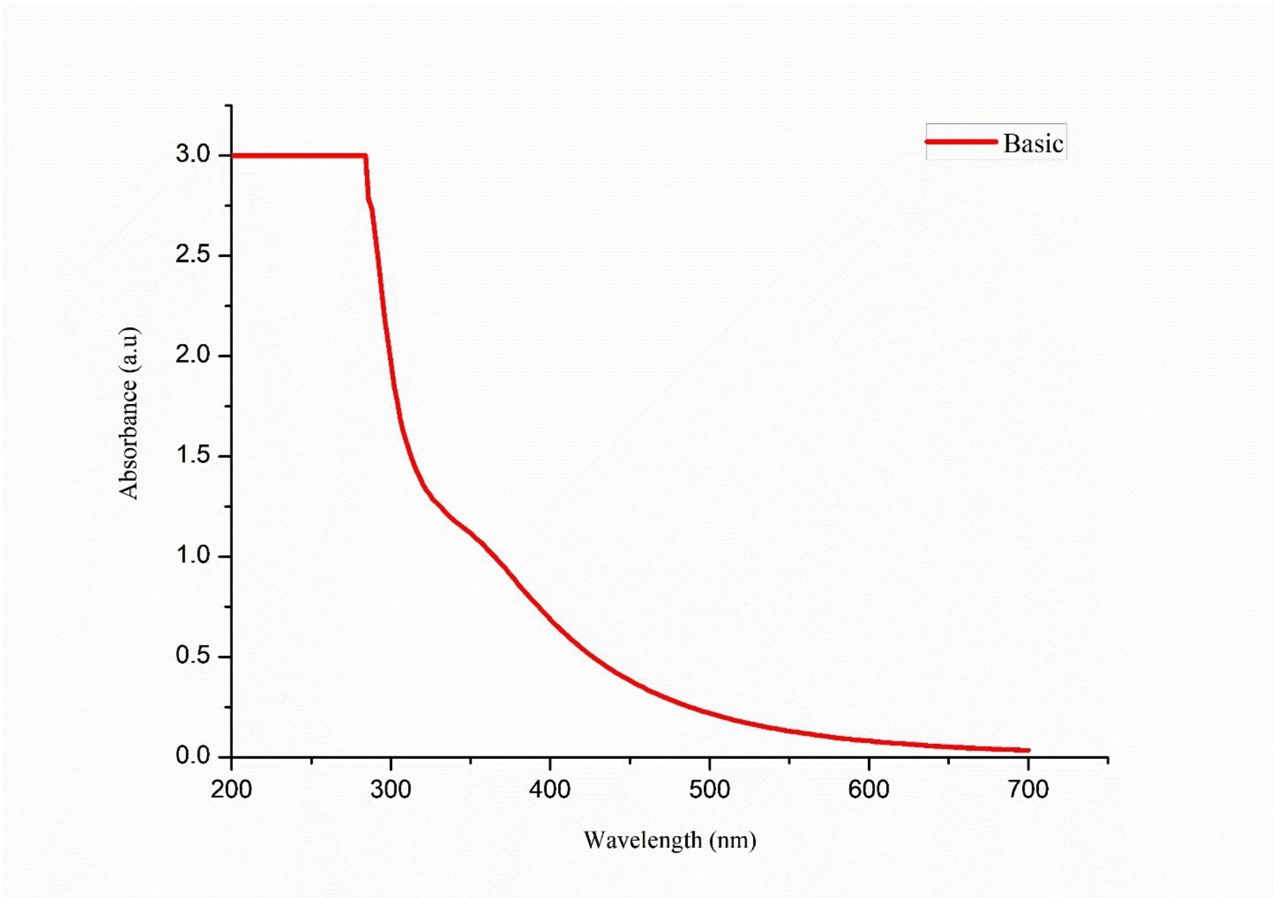

Fig: UV-Vis spectra of carbon dots in basic medium

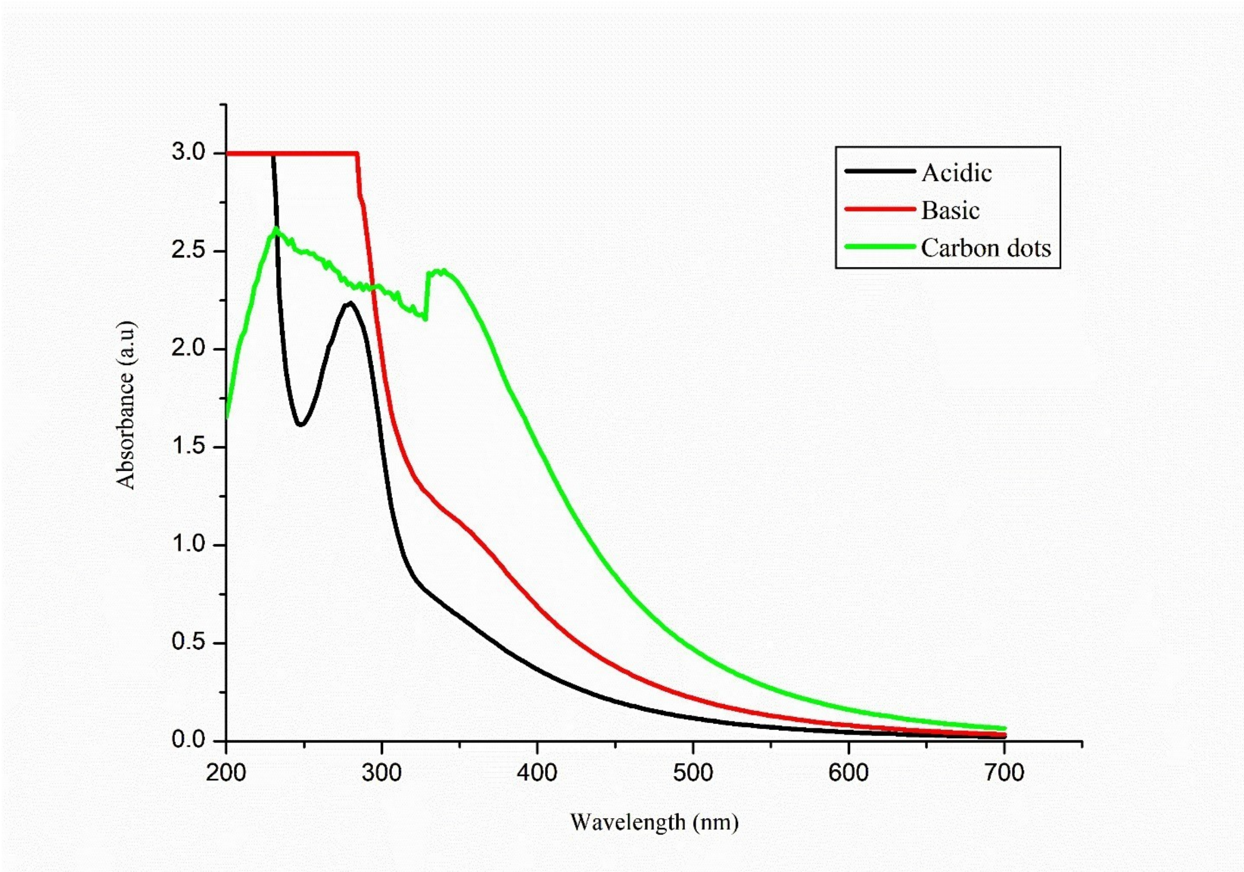

Fig: UV-Vis spectra of carbon dots alone and carbon dots in acidic and basic medium

The fluorescent carbon dots at different pH values are analysed and the absorption maxima in different pH is monitored. From the spectrum itself we can understand that significant changes in the absorbance are observed as the pH is varied. The colour of the solution changed from light yellow to dark brown as the pH increases. The result confirms that under low pH values, the carbon dots were dissolved as isolated species in the aqueous, in contrary, the aggregation of carbon dots appeared with increasing the pH value. The pH induced aggregation of the as prepared carbon dots results in an obvious fluorescence quenching at high pH value.These tunable fluorescence emission properties of as-prepared carbon dots were particular attractive for sensing, in vivo bio imaging and synthesizing novel photocatalysts etc.

CHAPTER 6

CONCLUSION

This study reports a green, cheap and convenient process for the synthesis of carbon dots via carbonization of glucose as the carbon precursor. Also they exhibit bright luminescence and their optical properties are pH dependent. These tunable properties of as synthesized carbon dots can be utilized for sensing, invivo bio imaging and synthesis of photo catalysts etc.

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