

## Research Article

# An Ultraviolet-visible Spectrophotometric Approach to Establish a Method for Determining the Presence of Fluorescein Sodium Dye

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## Abstract

This research look over the application of adsorbent as bentonite. Bentonite, aluminum phyllosilicate clay made up of montmorillonite. Montmorillonite known for enormous water absorption and high surface area. Dyeing industries are crucial ecological hazards donating upto twenty percent of industrial water pollution. These industries discharge toxic sewer water containing heavy metals, mutagenic substances, carcinogenic amines, and non-biodegradable chemicals, affects human health and aquatic ecosystems. The contamination of water with toxic dyes poses serious threats to surrounding. Therefore it is mandatory to overcome the environmental challenges with reliable and ability of component and environmental impact. Bentonite having capability to detoxified the harmful substances. It has the dormant for sorption the pesticide endrin possibly due to a combination of hydrophobic and charge-dipole. This study examined the potential of bentonite as adsorbent techniques for treating fluorescein sodium dyes from aqueous solutions. Experiment is done on the particular concentration of 200 ppm and at fixed pH of 2, 4 6, 8 and 10. Results recorded by the systronics spectrophotometer for absorbance Vs wavelength at pH 2 and 10 for fixed interval of time, Absorbance Vs Time, and decolorisation of dye. The performance of bentonite regarding color removal was optimum when the initial pH was 10 for 15 minutes and under ideal circumstances, decolorisation was shown to be 99.97% at alkaline pH. At pH 2 and 4 constant absorbance is achieved by 99.99% for a time intervals of 15 minutes. At an acidic pH the absorbance gradually decreased with respect to time. The spectrophotometric data at a wavelength of 490 nm is 2.683 at pH 10, shows the absorbance of toxic substances. As the wavelength is gradually increased the absorbance is also increased. The response of adsorbent as bentonite for the removal of fluorescein dyes from aqueous solution as well as for healthy environment is excellent and verified.

## Keywords

Fluorescein Sodium, Bentonite, Adsorbent, Decolorisation, Absorbance, Dye

## 1. Introduction

Without a doubt, the treatment of water, which is one of our most essential necessities, is critical [1]. The chemicals found in effluent from industrial activity have a harmful impact on

living organisms. Untreated textile effluents are discharged in the drain which deteriorate the water bodies [2-5]. Dyes such as MB, RhB, CR, OPD, PPD, Fluorescein Sodium, Disperse

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Violet 26, MR, and CV are the important sources of industrial pollutants originates from various industries such as the textile, cosmetic, leather, food, pharmaceutical, paint and varnish, and pulp and paper industries and used as colorants [6-9]. The textile industry, as a traditional manufacturing sector, is one of the industries with high labor intensity, large water consumption, and serious pollution. China is the largest exporter of textiles of all kinds, followed by the India, European Union and the USA. World's largest exporter of textiles, China's annual water consumption and wastewater discharge are staggered [10-13]. The American Dye Manufactures Institute stated that dye concentrations in coloured effluents ranges from 1000-1500 ADMI units. Composition of wastewater is extremely complicated, usually containing acids, bases, dyes, hydrogen peroxide, starch, surfactants, dispersants, and other chemical substances, and generally has a strong color and high concentration of organic compounds [14, 15]. Because of poor wastewater management policies of developing countries, 20% to 40% waterborne diseases are caused by industrial wastewater. At present one lakh chemical compounds are produced all over the world, with a total mass of approximately 70 thousand tonnes of dyestuff is manufactured yearly, and the majority of these dyes are organic as well as water soluble. Which results the bad biodegradability [16, 17]. The existence of dyes in the environment has mild to severe toxic effects on human health, including carcinogenic, mutagenic, allergic, and dermatitis effects, kidney disease and toxicity, causing serious harm not only to aquatic life but also to humans. As a result, an efficient dye removal method is imperative to minimize these issues [18].

Accordingly, countless studies have focused on effectively removing organic dyes from wastewater. The techniques utilized for the remediation of dye from wastewater using coagulation / flocculation, electrocoagulation, filtration, adsorption, ion-exchange, advanced oxidation processes (AOPs), activated sludge processes (ASP), sequencing batch reactors (SBR), membrane bioreactors (MBR), moving bed biofilm reactors (MBBR), Chemical Oxidation, Reverse Osmosis (RO), photo-Fenton, Biological Treatment and constructed wetlands (CW). Although these methods can be effective, they often involve high operating costs, the need to use chemical compounds that may be toxic, and in addition to, they can generate secondary pollution.

Throughout the adsorption method is efficient due to its low cost, ease of handling, and energy-saving features. Adsorption methods involve the movement of solids from the bulk liquid to the surface of the adsorbent. The adsorption of dyes is facilitated by hydrogen bonding and electrostatic interactions [19, 20]. The effectiveness of adsorption is determined both by the properties of the adsorbate (dye) and by the properties of the adsorbent—its specific surface area, volume and pore distribution, surface charge, functional groups, easy fabrication, high effective surface area, multi-functionalities, high surface volume ratio, high reactivity, large number of active sites, reusability, low cost, and high efficiency. Among the

countless materials tested as adsorbents of dyes from aqueous solutions are: activated carbons, zeolites, metal oxides, natural sorbents such as powdered walnut shells, and, more recently, biochars and bentonite.

Recent studies have been focused on the excellent adsorption capability of bentonite and have extensively utilized it as an adsorbent to remove organic dyes from wastewater. Bentonite has been used in its original form and it is sieved.

## 2. Materials and Methods

### 2.1. Materials

The chemicals utilized in this study, including fluorescein sodium dye (C. I. NO. 45350), M. W- 376.28, sodium hydroxide (98%), and hydrochloric acid (37%), were bought from Merck. UV-Visible Spectrophotometer 2703, equipped with a 1 cm (0.5 mL) quartz cell was utilized for recording the absorption spectrum and absorption measurements at a wavelength of 490 nm was employed. pH measurements were conducted using a Systronic digital pH meter with an integrated glass pH electrode. Micropipettes and volumetric flasks of appropriate sizes were employed to quantify the reagents and solution volumes. All measurements were carried out at the University department of Chemistry, India.

### 2.2. Methods

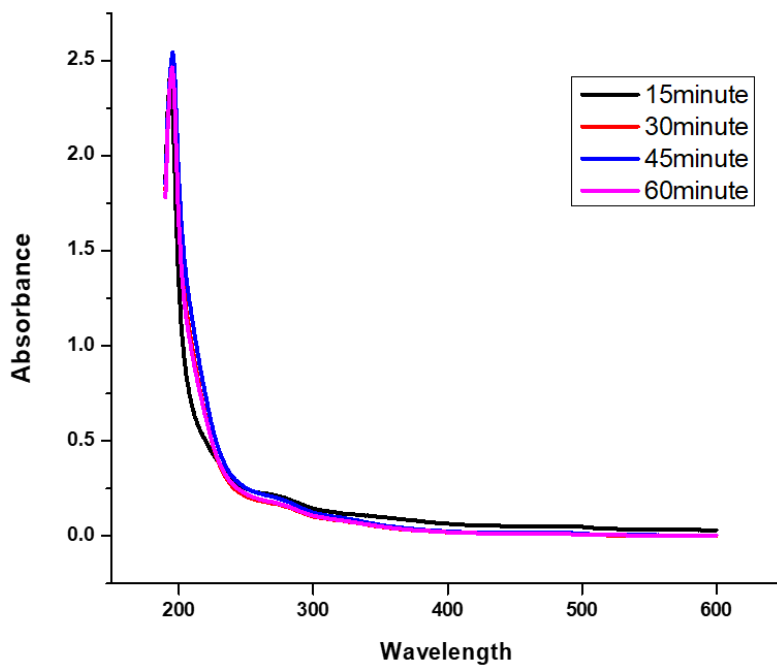
A fluorescein sodium dye stock solution was prepared by diluting the fluorescein powder with deionized water. Fluorescein sodium dye weighing was performed using a digital balance (SR NO. E-17496, Roy Electronics, Varanasi, India). A 0.2g dye was weighed and dissolved in 10 mL of deionized water in a 1000 mL volumetric flask. The volume was then made up to 1000 mL with deionized water to prepare a 200 ppm stock solution of 1000 mg/L fluorescein sodium dye. Take 100 ml of dye solution and measure the pH, weighed 1 g of bentonite and put in a 250 ml conical flask. Put this flask on the stirrer for proper mixing upto 15 minutes. Then filter the solution by whatman, 125 mm  $\phi$  filter papers. Similarly, experiment was done for fixed time intervals of 15 minutes and at pH of 2, 4, 6, 8 and 10. Filtrate were analysed by spectrophotometer AU 2703. Each dye was scanned in the wavelength ranging from 190 nm to 600 nm. The absorbance of the degraded sample was noted at  $\lambda_{max} = 490$  nm.

## 3. Results

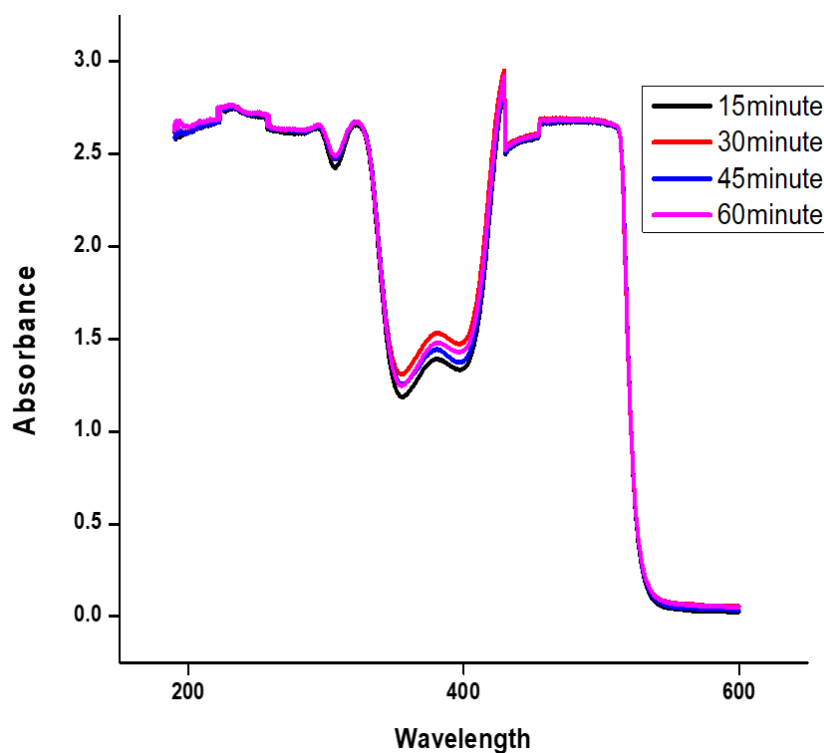
The fluorescein sodium dye absorbed on the surface of adsorbent i.e bentonite. The effect of pH on the adsorption of dye on adsorbent surface, was performed by varying the pH from 2 to 10 of dye solutions by using NaOH and HCl. The absorbance graph (Figure 1) at pH 2 shows that at a wavelength of 200 nm the maximum absorbance is 2.5 for 45 minutes. With

the increment of wavelength the absorbance is decreased. From, [Figure 2](#) it reveals that at higher pH 10 and concentration, the absorbance is maximum above 400 wavelength for 60 minutes. [Figure 3](#) depicts that with increase in time upto 60

minutes, at high concentration and basic medium, the absorbance capability of bentonite is maximum. [Figure 4](#) disclose the decolorisation percentage of dye by adsorbent at different time intervals.



*Figure 1. Absorbance at different wavelength at pH 2.*



*Figure 2. Absorbance at different wavelength at pH 10.*

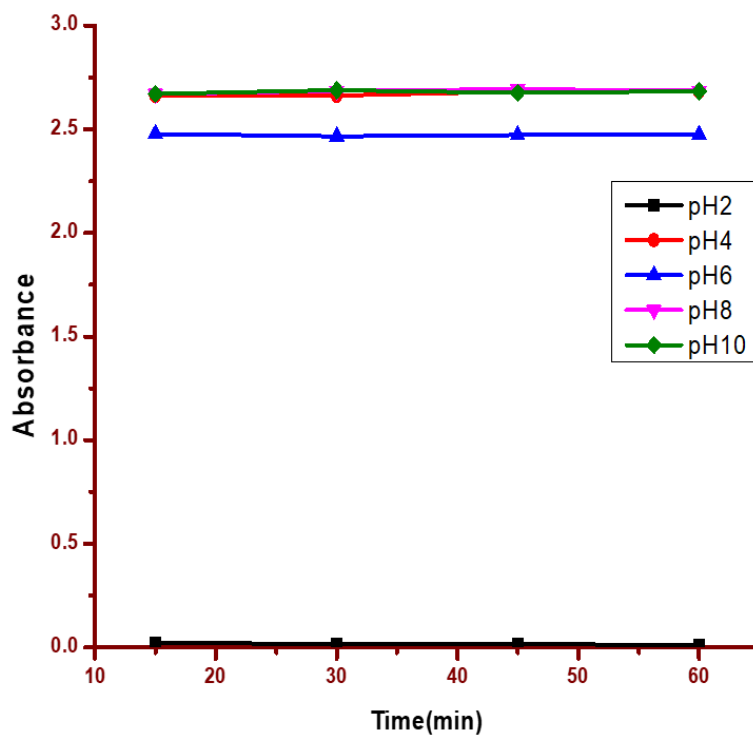


Figure 3. Absorbance at different time intervals and pH.

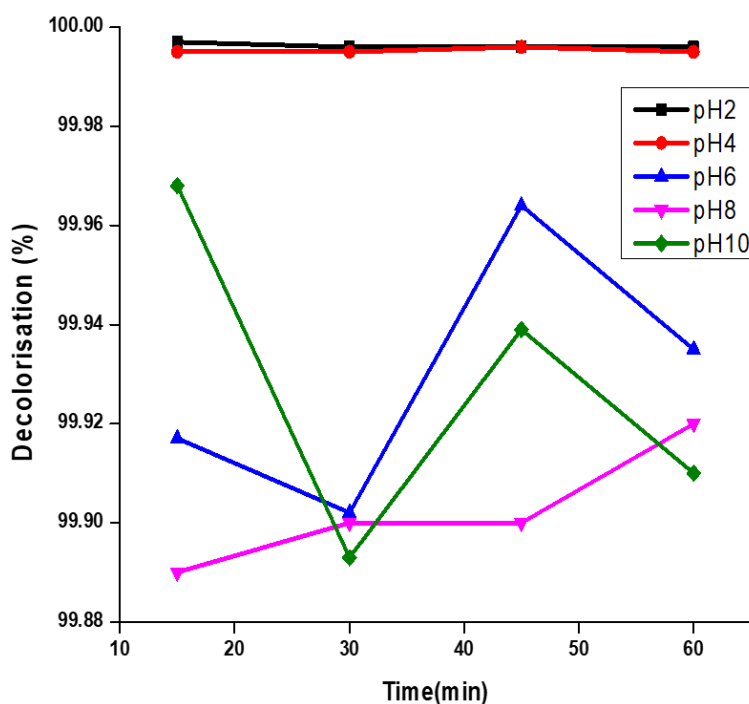


Figure 4. Decolorisation at different time intervals and pH.

The degradation rate was calculated from formula

$$\% \text{ decolorisation} = \frac{C_i - C_t}{C_i} \times 100$$

where,  $C_i$  stands for initial concentration and  $C_t$  stands for concentration at time  $t$ .

Table 1, Reveals the absorbance of fluorescein sodium dye at wavelength range from 400 nm to 500 nm, which is recorded by spectrophotometer. Fluorescein dye has a maximum absorbance wavelength of approximately 490 nm in aqueous solutions, producing a yellowish-green fluorescence. At this

wavelength the absorbance 2.683 was recorded. The absorbance is same upto 490.4 nm after 490.6 nm the absorbance is increased for a while and then gradually increase in wavelength results decrease in absorbance value upto 2.675.

*Table 1. Absorbance data for pH 10.*

Wavelength (nm)	Absorbance
500.0	2.675
499.8	2.675
499.6	2.677
499.4	2.676
499.2	2.678
499.0	2.678
498.8	2.679
498.6	2.679
498.4	2.679
498.2	2.680
498.0	2.680
497.8	2.681
497.6	2.681
497.4	2.682
497.2	2.681
497.0	2.681
496.8	2.681
496.6	2.682
496.4	2.684
496.2	2.685
496.0	2.685
495.8	2.684
495.6	2.683
495.4	2.681
495.2	2.681
495.0	2.681
494.8	2.682
494.6	2.683
494.4	2.684
494.2	2.683
494.0	2.681
493.8	2.679
493.6	2.678

Wavelength (nm)	Absorbance
493.4	2.679
493.2	2.682
493.0	2.683
492.8	2.683
492.6	2.683
492.4	2.683
492.2	2.684
492.0	2.682
491.8	2.681
491.6	2.681
491.4	2.684
491.2	2.685
491.0	2.685
490.8	2.684
490.6	2.684
490.4	2.683
490.2	2.683
490.0	2.683
489.8	2.684
489.6	2.685
489.4	2.686
489.2	2.684
489.0	2.684
488.8	2.683
488.6	2.684
488.4	2.684
488.2	2.685
488.0	2.684
487.8	2.683
487.6	2.683
487.4	2.683
487.2	2.684
487.0	2.686
486.8	2.687
486.6	2.688
486.4	2.687
486.2	2.687
486.0	2.686
485.8	2.686

Wavelength (nm)	Absorbance	Wavelength (nm)	Absorbance
485.6	2.685	477.8	2.687
485.4	2.684	477.6	2.686
485.2	2.684	477.4	2.686
485.0	2.683	477.2	2.685
484.8	2.684	477.0	2.684
484.6	2.684	476.8	2.685
484.4	2.685	476.6	2.685
484.2	2.684	476.4	2.686
484.0	2.683	476.2	2.683
483.8	2.683	476.0	2.683
483.6	2.682	475.8	2.682
483.4	2.683	475.6	2.684
483.2	2.684	475.4	2.684
483.0	2.685	475.2	2.686
482.8	2.686	475.0	2.686
482.6	2.686	474.8	2.686
482.4	2.686	474.6	2.685
482.2	2.687	474.4	2.684
482.0	2.686	474.2	2.684
481.8	2.685	474.0	2.683
481.6	2.685	473.8	2.684
481.4	2.684	473.6	2.685
481.2	2.684	473.4	2.687
481.0	2.686	473.2	2.688
480.8	2.687	473.0	2.689
480.6	2.687	472.8	2.688
480.4	2.687	472.6	2.687
480.2	2.686	472.4	2.687
480.0	2.686	472.2	2.686
479.8	2.685	472.0	2.686
479.6	2.684	471.8	2.686
479.4	2.685	471.6	2.687
479.2	2.686	471.4	2.685
479.0	2.688	471.2	2.685
478.8	2.689	471.0	2.685
478.6	2.690	470.8	2.687
478.4	2.689	470.6	2.688
478.2	2.688	470.4	2.688
478.0	2.687	470.2	2.688

Wavelength (nm)	Absorbance	Wavelength (nm)	Absorbance
470.0	2.687	462.2	2.688
469.8	2.686	462.0	2.688
469.6	2.686	461.8	2.684
469.4	2.687	461.6	2.683
469.2	2.690	461.4	2.680
469.0	2.691	461.2	2.681
468.8	2.691	461.0	2.682
468.6	2.690	460.8	2.683
468.4	2.690	460.6	2.683
468.2	2.689	460.4	2.682
468.0	2.688	460.2	2.681
467.8	2.687	460.0	2.681
467.6	2.687	459.8	2.681
467.4	2.686	459.6	2.684
467.2	2.685	459.4	2.685
467.0	2.685	459.2	2.685
466.8	2.684	459.0	2.684
466.6	2.683	458.8	2.684
466.4	2.682	458.6	2.685
466.2	2.683	458.4	2.687
466.0	2.682	458.2	2.687
465.8	2.684	458.0	2.686
465.6	2.683	457.8	2.683
465.4	2.685	457.6	2.683
465.2	2.686	457.4	2.683
465.0	2.687	457.2	2.683
464.8	2.688	457.0	2.682
464.6	2.686	456.8	2.682
464.4	2.687	456.6	2.683
464.2	2.687	456.4	2.684
464.0	2.686	456.2	2.684
463.8	2.684	456.0	2.681
463.6	2.683	455.8	2.681
463.4	2.685	455.6	2.678
463.2	2.685	455.4	2.680
463.0	2.685	455.2	2.669
462.8	2.685	455.0	2.655
462.6	2.686	454.8	2.626
462.4	2.689	454.6	2.612

Wavelength (nm)	Absorbance	Wavelength (nm)	Absorbance
454.4	2.604	446.6	2.591
454.2	2.603	446.4	2.591
454.0	2.603	446.2	2.590
453.8	2.601	446.0	2.589
453.6	2.600	445.8	2.588
453.4	2.600	445.6	2.589
453.2	2.601	445.4	2.589
453.0	2.602	445.2	2.589
452.8	2.602	445.0	2.589
452.6	2.601	444.8	2.588
452.4	2.601	444.6	2.588
452.2	2.600	444.4	2.587
452.0	2.600	444.2	2.586
451.8	2.599	444.0	2.587
451.6	2.599	443.8	2.586
451.4	2.598	443.6	2.585
451.2	2.598	443.4	2.584
451.0	2.597	443.2	2.583
450.8	2.598	443.0	2.583
450.6	2.598	442.8	2.583
450.4	2.598	442.6	2.582
450.2	2.597	442.4	2.581
450.0	2.598	442.2	2.581
449.8	2.598	442.0	2.581
449.6	2.597	441.8	2.580
449.4	2.597	441.6	2.579
449.2	2.596	441.4	2.579
449.0	2.596	441.2	2.579
448.8	2.595	441.0	2.578
448.6	2.594	440.8	2.577
448.4	2.594	440.6	2.576
448.2	2.594	440.4	2.577
448.0	2.594	440.2	2.575
447.8	2.594	440.0	2.575
447.6	2.594	439.8	2.574
447.4	2.593	439.6	2.574
447.2	2.592	439.4	2.572
447.0	2.591	439.2	2.571
446.8	2.591	439.0	2.570

Wavelength (nm)	Absorbance	Wavelength (nm)	Absorbance
438.8	2.569	431.0	2.529
438.6	2.568	430.8	2.526
438.4	2.568	430.6	2.524
438.2	2.569	430.4	2.521
438.0	2.569	430.2	2.554
437.8	2.568	430.0	2.611
437.6	2.567	429.8	2.753
437.4	2.566	429.6	2.848
437.2	2.565	429.4	2.920
437.0	2.564	429.2	2.918
436.8	2.563	429.0	2.911
436.6	2.562	428.8	2.903
436.4	2.561	428.6	2.893
436.2	2.560	428.4	2.886
436.0	2.560	428.2	2.877
435.8	2.560	428.0	2.870
435.6	2.558	427.8	2.859
435.4	2.558	427.6	2.850
435.2	2.558	427.4	2.839
435.0	2.557	427.2	2.833
434.8	2.555	427.0	2.825
434.6	2.553	426.8	2.819
434.4	2.552	426.6	2.809
434.2	2.552	426.4	2.798
434.0	2.551	426.2	2.787
433.8	2.551	426.0	2.775
433.6	2.550	425.8	2.764
433.4	2.550	425.6	2.755
433.2	2.549	425.4	2.745
433.0	2.546	425.2	2.737
432.8	2.545	425.0	2.724
432.6	2.542	424.8	2.712
432.4	2.542	424.6	2.696
432.2	2.541	424.4	2.684
432.0	2.540	424.2	2.669
431.8	2.539	424.0	2.657
431.6	2.537	423.8	2.644
431.4	2.534	423.6	2.632
431.2	2.531	423.4	2.620

Wavelength (nm)	Absorbance	Wavelength (nm)	Absorbance
423.2	2.608	415.4	1.988
423.0	2.594	415.2	1.973
422.8	2.580	415.0	1.958
422.6	2.564	414.8	1.943
422.4	2.548	414.6	1.929
422.2	2.533	414.4	1.915
422.0	2.518	414.2	1.901
421.8	2.502	414.0	1.887
421.6	2.487	413.8	1.874
421.4	2.471	413.6	1.861
421.2	2.456	413.4	1.849
421.0	2.440	413.2	1.836
420.8	2.425	413.0	1.824
420.6	2.409	412.8	1.812
420.4	2.393	412.6	1.800
420.2	2.377	412.4	1.788
420.0	2.361	412.2	1.776
419.8	2.345	412.0	1.764
419.6	2.328	411.8	1.753
419.4	2.312	411.6	1.742
419.2	2.295	411.4	1.732
419.0	2.280	411.2	1.721
418.8	2.264	411.0	1.711
418.6	2.247	410.8	1.701
418.4	2.231	410.6	1.691
418.2	2.214	410.4	1.681
418.0	2.198	410.2	1.672
417.8	2.182	410.0	1.663
417.6	2.166	409.8	1.654
417.4	2.149	409.6	1.645
417.2	2.133	409.4	1.636
417.0	2.117	409.2	1.628
416.8	2.101	409.0	1.620
416.6	2.085	408.8	1.611
416.4	2.067	408.6	1.604
416.2	2.051	408.4	1.596
416.0	2.034	408.2	1.589
415.8	2.019	408.0	1.583
415.6	2.003	407.8	1.576

Wavelength (nm)	Absorbance
407.6	1.570
407.4	1.563
407.2	1.557
407.0	1.551
406.8	1.545
406.6	1.540
406.4	1.534
406.2	1.529
406.0	1.524
405.8	1.519
405.6	1.515
405.4	1.510
405.2	1.505
405.0	1.499
404.8	1.493
404.6	1.489
404.4	1.484
404.2	1.481
404.0	1.477
403.8	1.474
403.6	1.471
403.4	1.468
403.2	1.465
403.0	1.462
402.8	1.460
402.6	1.457
402.4	1.455
402.2	1.453
402.0	1.451
401.8	1.449
401.6	1.447
401.4	1.446
401.2	1.444
401.0	1.442
400.8	1.441
400.6	1.440
400.4	1.439
400.2	1.437

Wavelength (nm)	Absorbance
400.0	1.437

## 4. Discussion

Figure 1. Depicts that the acidic behaviour of fluorescein dye at pH 2. The maximum absorbance is 0.021 at 15 minutes for 200 ppm at a wavelength of 490 nm. As the wavelength linearly increased, the absorbance continuously decreased upto 0.004 at 600 nm. Whereas at alkaline pH 10, the absorbance is 2.671 for 15 minutes, 2.688 for 30 minutes, 2.676 for 45 minutes and the maximum absorbance is 2.683 achieved at a time interval of 60 minutes at 490 nm as shown in Figure 2.

Figure 3 exhibit that at an acidic pH 2 the absorbance is poor but at an alkaline pH 10 and time interval of 60 minutes the absorbance is maximum for 200 ppm.

Figure 4. Clearly shows that at acidic pH 2 and 4 and higher concentration the approximately the constant decolorisation percentage is achieved by 99.99% for a time interval of 15 minutes. At neutral pH 8, the maximum decolorisation percentage, 99.96% is achieved with linearly increase in time.

At pH 10, initially the decolorisation is increased and decolorisation percentage 99.97 is recorded, after 15 minutes it decreased and then again increased for a while. At pH 6, the decolorisation percentage is continuously increased upto 45 minutes and then decreased.

## 5. Conclusions

This study demonstrates the bentonite performance, easy availability, cost effectiveness, high porosity, recycling ability, and abundant active sites on its surface, as bentonite was used for the removal of fluorescein sodium dye. At acidic pH the constant decolorisation percentage is achieved by 99.99% for a time interval of 15 minutes. At pH 10 the initial decolorisation percentage is 99.98 but after a time interval of 15 inutes it decreased.

## Abbreviations

MB	Methylene Blue
RhB	Rhodamine B
CR	Congo Red
OPD	Orthophenylenediamine
PPD	Paraphenylenediamine
DV	Disperse Violet
MR	Methyl Red
CV	Crystal Violet

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## Author Contributions

**Amit Anand:** Conceptualization, Data curation, Formal Analysis, Investigation, Methodology, Writing – original draft

## Data Availability Statement

The data supporting the outcome of this research work has been reported in this manuscript.

## Conflicts of Interest

The author declares no conflicts of interest.

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## Biography



**Amit Anand**, Research Scholar Tilka Manjhi Bhagalpur University, Bhagalpur, India. My research work is based on dyes and water pollution. My Five research articles are published in different peer reviewed and scopus journals. I am honoured with various prestigious awards such as best paper presentation (2026) by Ixora Research Association, Jharkhand, India; Best Young Researcher Award-2023 by Global Leader Foundation, Delhi, India and Indian Icon Awards (2024) by Kiteskraft Productions LPP, Punjab, India. I believes that “learning is not about remembering facts, it’s about thinking and investigating how things work.”