

## Effect of He-Ne Laser (632.8 Nm) in Fluorescence Efficiency of Solvents

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### ABSTRACT

*In this work, the laser of He-Ne absorbed by beet dyes makes effects of the dyes, which studied as PH effect. The dyes solution was separated by using tow technique; the HPLC-absorption and HPLC fluorescence techniques to get information about the number of the components present in the dye solution and both techniques give the same result indicating the presence of four components.*

*The dye solutions were pumped by He-Ne laser (632.8 nm) and the fluorescence efficiency was displayed by (usb 2000 spectrophotometer detector). the proportional relation between absorbance and fluorescence was investigated in this study.*

**Keywords:** He-Ne laser; Solvents; absorbance; fluorescence

### INTRODUCTION

Now days, dye lasers are used in many applications, including spectroscopy, communication, industry, holography, and also in medical. A laser is a device that emits light with specific properties through a process called stimulated emission.[1,2,3]

A variety of environmental factors affect fluorescence emission, including interactions between the fluorophore and surrounding solvent molecules (dictated by solvent polarity), other dissolved inorganic and organic compounds, temperature, pH, and the localized concentration of the fluorescent species. The effects of these parameters vary widely from one fluorophore to another, but the absorption and emission spectra, as well as quantum yields, can be heavily influenced by environmental variables. In fact, the high degree of sensitivity in fluorescence is primarily due to interactions that occur in the local environment during the excited state lifetime. This interactive tutorial explores relaxation effects and associated spectral shifts that occur as a function of solvent polarity.[4]

### ABSORPTION AND FLUORESCENCE

**Table 1. Commercial dye laser output characteristics**

<i>Pump Source</i>	<i>Argon / Krypton Cw</i>	<i>Ion Laser Mode- Locked/Cavity Dumped</i>	<i>Flash Lamp Laser</i>	<i>Excimer Laser</i>
Tuning Range [nm]	380-950	580-880	335-850	320-1,024
Average Power [W]	5	0.1	3	10
Repetition Rate Hz]	CW	3.8M	2-30	1-500
Peak Power [kW]	-----	10	7,000	-----
Energy/pulse [mJ]	-----	0.01	3,500	40-120
Pulse width [nsec]	-----	0.0001	260-600	7-250
Line width [GHz]	0.0005 to 40	Various	2	0.15

Dye lasers can be pumped by incoherent or laser sources, both pulsed and continuous wave (CW), and offer the possibilities of broad wavelength control, multi joule pulsed operation, ultra narrow line-widths, or ultra-short pulses as shown in table (1). [5]

A photon can be absorbed by the dye molecule in the lowest vibration sublevel  $a$  of the  $S_0$  ground state, producing transitions to one of the upper vibration levels of  $S_1$  denoted by  $B$ . The molecule then undergoes a radiation less decay,  $B \rightarrow b$ , to the bottom of  $S_1$ .

The radiation less transition involves a rearrangement of the total energy within a system without photon emission. In most instances, the energy is absorbed in increased kinetic energy or motion of the atoms in the system and appears as heat. After some finite time, given by the excited state lifetime of  $S_1$ , the molecule may decay spontaneously to the lower energy level  $S_0$ , conserving energy in the process with the release of a photon. This is known as spontaneous emission in which the photon energy equal to the energy difference  $E_b - E_A$  between the states in which the transition occurs. The wavelength is given by; [6,7,8]

$$\lambda_{bA} = \frac{hc}{E_b - E_A} \dots\dots\dots (1)$$

Where  $h$  = blank constant and  $c$  = light speed . Another radiation less transition,  $A \rightarrow a$ , returns the excited molecule to its ground state.

$$\lambda_{aB} = \frac{hc}{E_a - E_B} \dots\dots\dots (2)$$

Notice that the wavelength of light emitted by the dye when it fluoresces is longer (lower energy) than that of the absorbed light:

$$E_b - E_A < E_B - E_a \xrightarrow{\text{yields}} \lambda_{aB} < \lambda_{bA} \dots\dots\dots (3)$$

This property of organic dyes is used in a number of familiar commercial products such as brightly colored plastics and household detergents where certain dyes act as brightening (whitening) agents by absorbing UV and emitting in the blue portion of the spectrum.

The ability to absorb light of short wavelengths and fluoresce or re-emit at longer wavelengths is a useful property of organic dyes.

Most of photochemistry is concerned with molecules in the condensed phase, i.e. in liquids or solids, and intermolecular solute - media interactions affects the energy of the electronic states. As the interaction energy depends on the nature and the properties of the respective state, it will be different for ground and excited state molecules and gives thence rise to spectral shifts, normally referred to as solvaochromic shifts. Solvetochromy is thus an excellent measure for variations of the relative energies of the molecular states in different environments.

With the exception of water, all solvents should be considered hazardous. In many instances, the solvent in which the dye is dissolved plays a major role in the hazard presented by the final solution.

These hazards must be addressed carefully in dye handling and solution preparation. Nearly all solvents are highly flammable. Therefore, a small fire extinguisher should be installed near the laser in a readily accessible and unobstructed area.

A particular fire hazard that is not commonly known occurs with non-polar and, hence, nonconductive solvents. If these solvents are circulated at a high speed through plastic tubing's, the pump unit acts as a van de Graff generator, producing up to 100 kV, and sparks may pierce the tubing and ignite the solvent. The dye selector's use grounding wires inside the plastic tubing's to eliminate these problems. However, when using such solvents, check

first for static electricity before opening the reservoir. Static electricity is present when hair on the back of your hand or forearm is attracted to the plastic tubing. Do not circulate dye solutions made with such solvents for more than a minute, unless the cuvette has been placed into the crate and is grounded.[9]

Alkaline conditions cause aldimine bond hydrolysis, while acidification induces re-condensation of betalamic acid with the amine group of the addition residue, which seen in Fig. (1)[10]

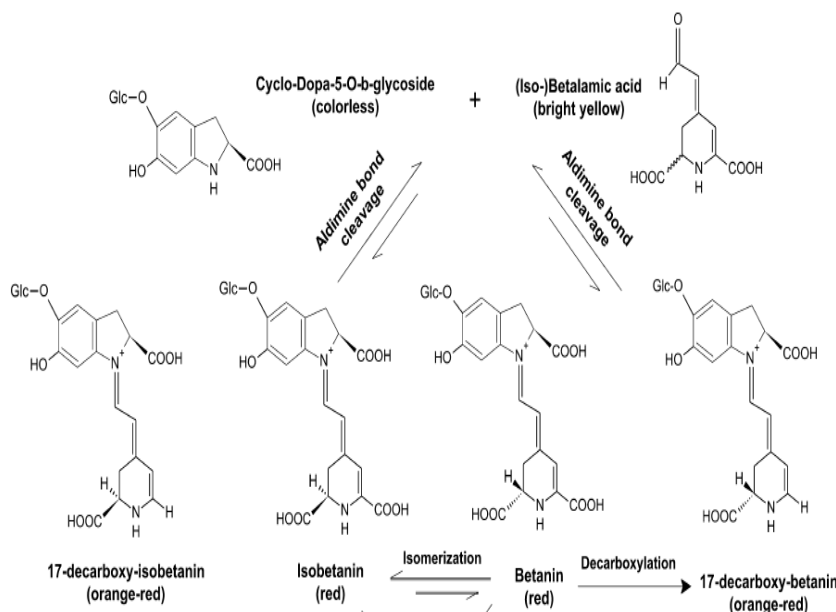


Figure 1. Beet dyes in acidic and alkaline conditions

## PREPARATION SAMPLE

The beet samples were collected from local markets in Khartoum state and the dye solution was extracted by water and the extracted was separated by filtration. The dye solution was extracted by 0.1 % aqueous TFA (Tetra FluoroAcetic acid). **The water added to the solutions to obtain four samples of various concentrations as;**

**Sample (1):** 1ml of dye 0.1 solution was added to 100ml of water (the absorbance of sample was adjusted to be less than 0.1au. Four different concentrations were prepared by water dilution in the range of (0.1 - 0.01 au abs).

**Sample (2):** 1ml of dye solution was added to 100ml of glycerin –water (1: 10) solution. Four different concentrations were prepared by dilution in the range of (0.1 -0.01au abs).

**Sample (3):** 1ml of dye solution was added to 100ml of glycerin – water(2: 10) solution. Four different concentrations were prepared by dilution in the range of (0.1 – 0.01 au abs).

**Sample (4):** 1ml of dye solution was added to 100ml of glycerin – water (4:10) solution. Four different concentrations were prepared by dilution in range of (0.1 -0.01 au abs).

The effect of pH on the absorption of light can be studied by PH meter, uv-vis spectrophotometer, hydrochloric acid (0.01 and 0.001M) or Sodium hydroxide (0.01 and 0.001M)

Samples were prepared by water extraction from thin slices of fresh beet root (100 gm of slices doped in 500 ml of distilled water for 48 hours at room temperature), the pH of samples

adjusted by addition of hydrochloric acid and sodium hydroxide the different pH samples were placed in uv-visible spectrophotometer and the values were recorded. The components of dyes solution separated by tow process as:

First, the process was carried out on a LC- 10 AT-VP chromatograph with a photodiode array waters 2996 detector equipped with a LiChoro Cart250 – 4.6, Li Chrospher 100RP-18 (5µ m) Column by used HPLC absorption .

Second, process was carried out on a LC – 10AT – VP chromatograph with a fluorescence RF. 10 a sykmn detector equipped with a LiChoro Cart 250 -4.6, LiChrospher 100 RP- 18 (5µ m) Column by used HPLC fluorescence.

In all process, the separation was preformed isocratically using a mixture of 90% solvent A (0.5%aqueous TFA) with 10 %solvent B (acetonitrile) for 35 min at a flow rate of 0.1 ml /min (injection volume10µL).

## MATERIAL AND METHOD

In this paper, tunable He – Ne laser (110 /220 VAC, 50 – 60 Hz), 22.0mW minimum 24.0mW typical 27.0mW maximum, (632.8 nm) linear Polarization 500:1, TEM<sub>00</sub>, 0.80 mm Beam Diameter was used to absorbed by the samples. Usb 2000 spectrophotometer specification detector range of (200 -1100) nm, entrance aperture(5,10,25,50,100,200)mm wide slits or fiber, focal length [f/4,42mm(input) ;68mm(output)], sensitivity (estimate) of (400nm – 90, 600nm – 41, 800nm – 203) photons /count, fiber optic connector SMA 905 to single – strand optical fiber (0.22NA), data transfer rate; full scans into memory every 13 milliseconds, integration time of (0.003 to 65) seconds, produced by Ocean optics, USA), used to measure and record the fluorescence comes of samples, which placed in quartz cell , dye solutions of various concentrations (samples) were studied.

Dye solution of samples and the solvent of samples were placed in quartz cell and pumped by He –Ne laser the fluorescence of samples was recorded by using usb 2000 spectrophotometer (ocean optics USA).For all spectra measurement, the emission was collected at 90 ° from the excitation beam as shown in the Fig (2).

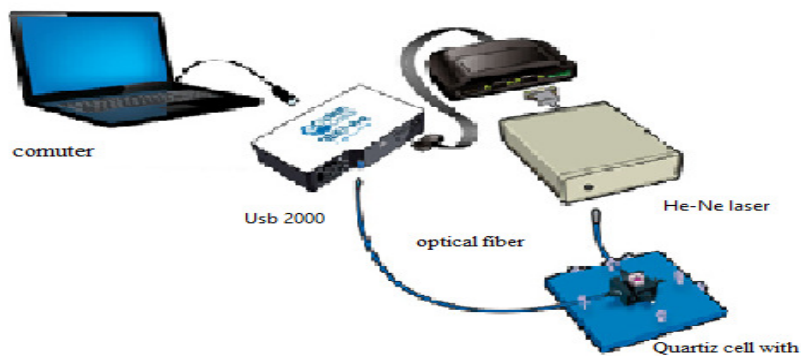


Figure 2. Experimental setup used to measure the emission spectra of samples and solvent

## RESULTS OF THE PH AFFECTION ON $\lambda_{MAX}$ FOR DYE SOLUTION

Result of absorption of beet dyes on different pH (3-12) show that the  $\lambda_{max}$  of light absorption is shifted and the dye solution showed instability at pH more than 8 as shown in the figures (3-11).

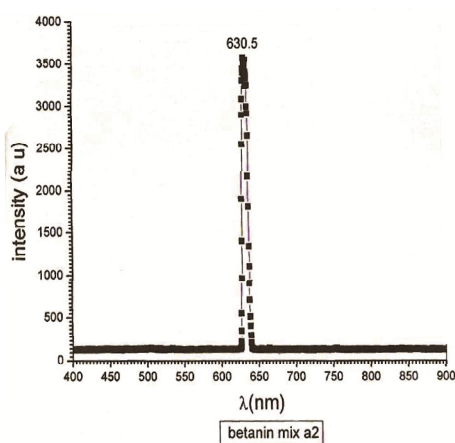


Figure 3. Fluorescence spectra of water extracted C2

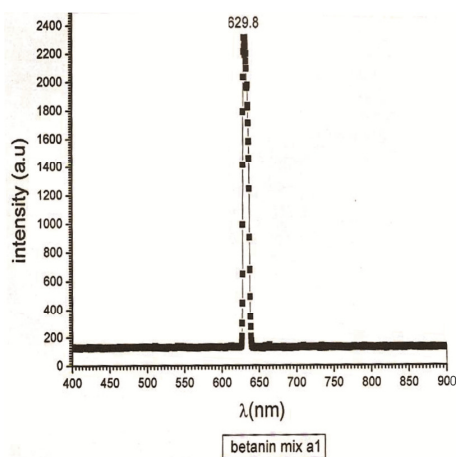


Figure 4. Fluorescence spectra of water extracted C1

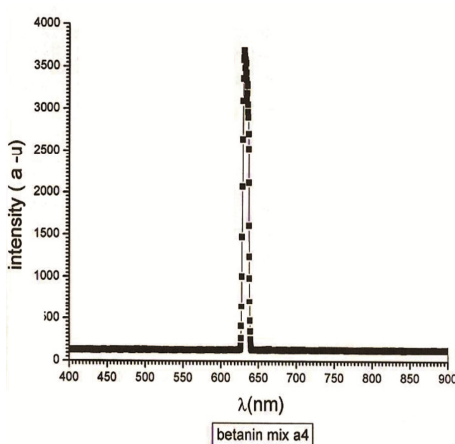


Figure 5. Fluorescence spectra water extracted C4

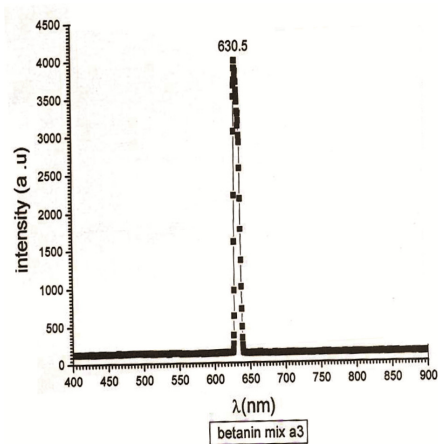


Figure 6. Fluorescence spectra water extracted

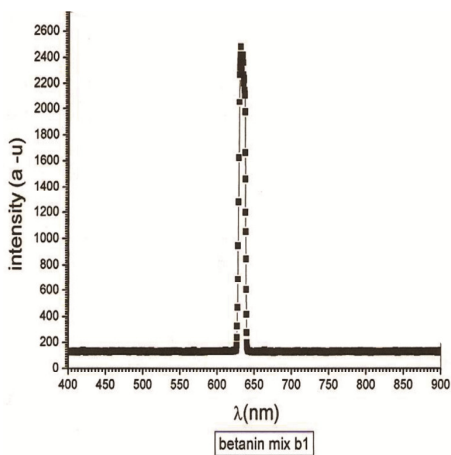


Figure 7. Fluorescence spectra of Ethanol extracted C2

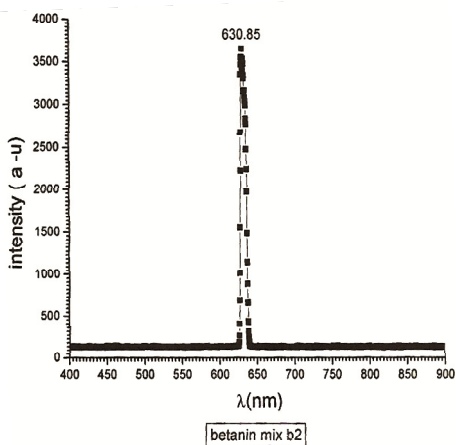


Figure 8. Fluorescence spectra of Ethanol extracted C1

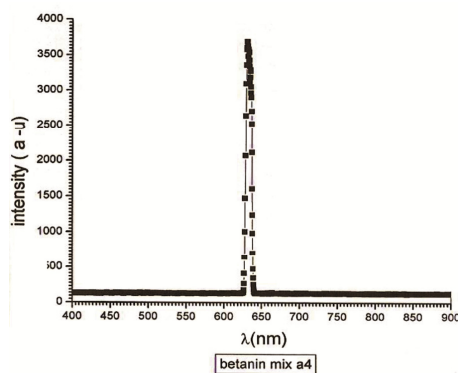


Figure 9. Fluorescence spectra of Ethanol extracted C4

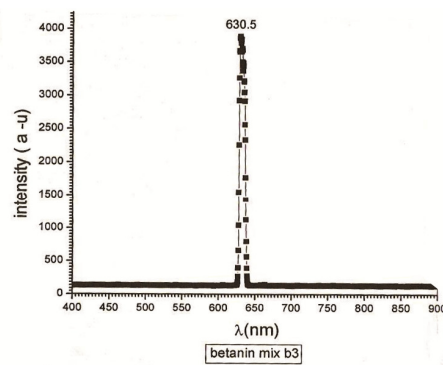


Figure 10. Fluorescence spectra of Ethanol extracted C3

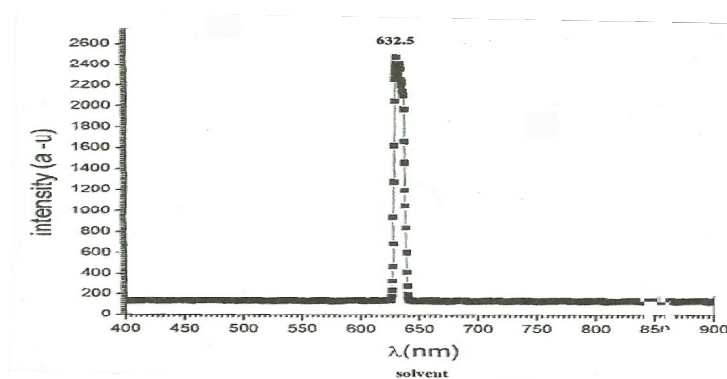


Figure 11. Fluorescence spectra of solvent

Result of HPLC- fluorescence for beet dyes (running in 530 nm excitation wave length and the emission wave length at 630 nm).

**RESULTS MEASURED BETWEEN ABSORBANCE AND FLUORESCENCE**

The result which measured between absorbance and fluorescence for the four samples (1, 2, 3, 4 and standard) were tabled on the Table labeled as 2, 3, 4, 5, and 6 respectively.

The relation between absorbance and fluorescence for the four samples (1, 2, 3, 4 and standard), were shown by figures (12, 13, 14, 15, and 16), respectively.

**Table 2. Abs-flu sample 1**

Absorbance(au)	0.015	0.046	0.066	0.084
Fluorescence(au)	5.12	13.68	22.72	28.03

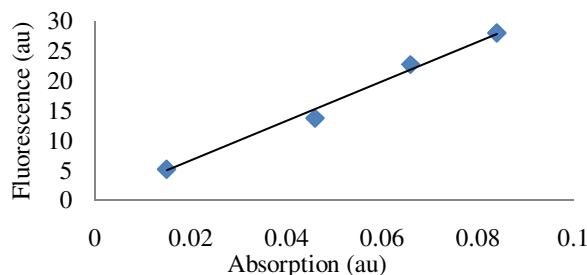


Figure 12. Shows the absorbance versus fluorescence of sample (1)

**Table 3. Abs-flu sample2**

Absorbance (au)	0.016	0.043	0.067	0.084
Fluorescence(au)	8.56	20.93	32.83	39.96

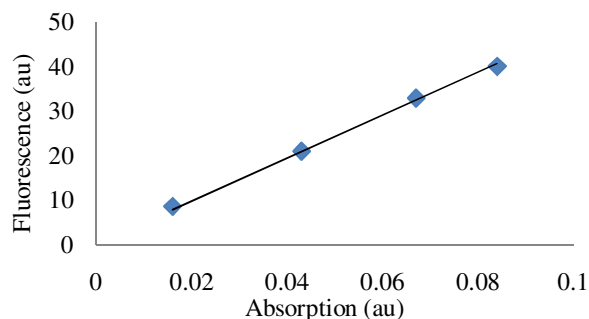


Figure 13. Shows the absorbance versus fluorescence of sample (2)

**Table 4. Abs-flu sample 3**

Absorbance(au)	0.018	0.048	0.069	0.086
Fluorescence(au)	11.82	26.55	37.72	48.33

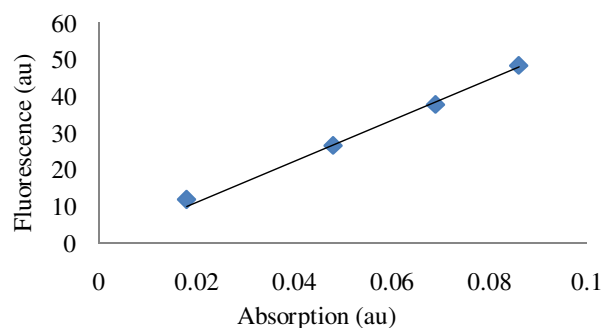


Figure 14. Shows the absorbance versus fluorescence of sample (3)

**Table 5. Abs-flu sample 4**

Absorbance(au)	0.021	0.046	0.070	0.086
Fluorescence(au)	22.19	39.98	60.81	76.45

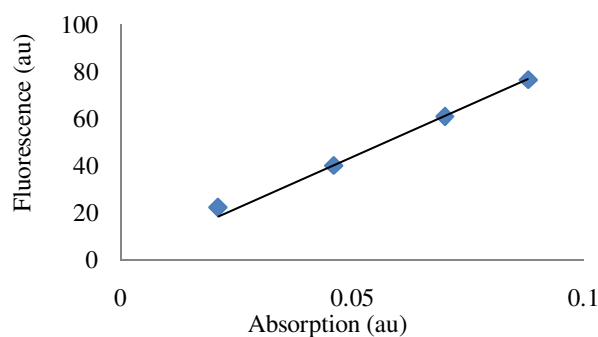


Figure 15. Shows the absorbance versus fluorescence of sample (4)



**Table 6. Abs-flu standard**

Absorbance(au)	0.022	0.041	0.062	0.082
Fluorescence(au)	19.85	36.06	55.92	73.96

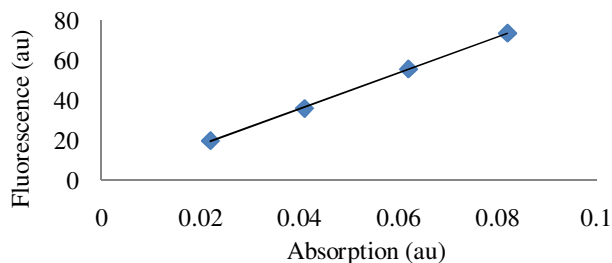


Figure 16. Shows the absorbance versus fluorescence of standard

## DISCUSSION

On the basis of the results presented in this work, it can initially be concluded that lasing in beet dyes solution is possible. This is in agreement with the results of fluorescence of beet dyes solutions. The measurements showed that the lasing wavelength depends on the pH of the solvent. Also in this study the same results indicated by both techniques (HPLC-absorption and HPLC fluorescence). The absorbance and fluorescence increase with increasing of concentration of the sample was investigated as seen in figures (12-15). Also these figures had shown the proportional relation between absorbance and fluorescence of all samples.

The  $\lambda_{\max}$  of light absorption is shifted as a result of the pH varying and the dye solution showed instability at pH more than 8.

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