Investigation of third order nonlinearity of Ethidium bromide doped deoxyribonucleic acid (DNA)

R. K. FakherAlfahed¹, Hussain Ali Badran², Abu Talib Y. Abbas¹, Noor Al-Huda Saleh⁴
¹Al-Nahrain University, Al-Nahrain Renewable Energy Research Center, Baghdad, Iraq.
²Basrah University, Education College for Pure Sciences, Physics Department, Basrah, Iraq.
³Education Ministry, General Directorate of Education, Qamar Bani Hashem Intermediate School, Basrah, Iraq.
⁴Ministry of Transport, General Company of Iraq Ports, Ports institute, Basrah, Iraq.

Corresponding author E-mail: r.k.fakheralfahed@nerc.nahrainuniv.edu.iq

Abstract. The concentrations-dependent refractive index $n_2$ and the nonlinear absorption coefficient $\beta$ of Ethidium bromide dye-doped deoxyribonucleic acid (biological polymer DNA) solutions in the SDL regime at 532 nm are reported. The Z-scan technique was performed in two ways and two different wavelengths, 532nm and 473nm, the open aperture technique and the closed aperture technique. From open aperture Z-scan measurements it is found that the Ethidium bromide doped deoxyribonucleic acid films exhibited reverse saturable absorption. The coefficient of nonlinear refraction and nonlinear absorption coefficient at 473nm wavelength is greater than at 532nm.

key word: Ethidium bromide, Reverse saturable, Laser , DNA.

1. Introduction

During the last decade, dye doped deoxyribonucleic acid (DNA) or polymer are being focused because of their technological applications in optical devices [1-3], spintronics, human eyes [4], solar cells [5], catalysis, optical and gas sensors protection [6], all optical switching [7], holographic gratings [8], optical storage [9] and super capacitors [10]. The characteristic feature of a deoxyribonucleic acid biopolymer is rod-like, double helix with $\pi$-electron-rich base pair stacking through hydrogen bonds between the bases and are stabilized by $\pi$-$\pi$ interactions [11]. However, azo dye (Ethidium bromide) is a good material to be used as optical limiting materials. Many dyes can be easily inserted into the grooves of deoxyribonucleic acid helix. Some of other azo dyes, optical dyes or organic materials can be easily stacked on the surface of deoxyribonucleic acid helix. Since early work of doping dye into deoxyribonucleic acid (DNA) polymer [12,13], great investigators have demonstrated the nonlinear optical coefficients, optical limiting properties and self diffraction pattern of dye doped deoxyribonucleic acid matrices. Nonlinear optical (NLO) properties of deoxyribonucleic acid biopolymer in solution form [14], in azo dye films [15], in organic compounds [16] and in Rhodamine 6G-PVA [17] has been investigated recently. The mechanism of operation of this technique is based on the principle of spatial beam distortion [18], which grows from the optically induced nonlinear refractive index. From this method one can obtain both the nonlinear signal, the magnitude of the nonlinearity and the nonlinear refractive index value easily from experimental readings with a simple readout analysis [19]. Another benefit of this technique is its ease of application in studying transient phenomena [20]. The studies of Z-scan technique not only provide information about the non-linear optical properties of materials, but also provide important information regarding response time [21] and dynamics of transient processes, which contribute to obtaining the nonlinear refractive index [22].
In this paper, we employ the close and open Z-scan technique to study the nonlinear refractive index nonlinear absorption property of Ethidium bromide dye doped deoxyribonucleic acid (DNA) biopolymer, the nonlinear coefficients response of EtBr dye doped biopolymer was measured with 532 nm and 473 nm CW laser.

2. Experimental measurements

2.1 Samples preparation

Using a sensitive digital scale, about 0.0315g of EtBr dye powder was dissolved in 10 ml of distilled water. A concentration of 8 mM was obtained, after which the 8 mM main solution was placed on the heating mixer for 30 minutes at a 45 °C temperature of enough degree to ensure that the dissolution is complete. Then, the filtration process was carried out in two ways, the first with filter paper, and followed directly by filter syringe with a thickness of 0.2 μm. After completing these processes, we obtain a completely dissolved, clear solution. The biological polymer DNA was doped with concentrations of (1, 2, 3 and 4 mM). A polymer solution sample of each concentration is placed in a quartz cell, the cell thickness is 1mm and the absorbance of the stain-stained DNA polymer solution is measured. Fig. 1a and b show the chemical structure and molecular formula for Ethidium bromide dye and deoxyribonucleic acid (DNA), respectively.

![Chemical structure of (a) EtBr dye and (b) biological polymer deoxyribonucleic acid (DNA).](image)

2.2 Absorption measurement

The UV–visible absorption spectra of the EtBr dye doped deoxyribonucleic acid (DNA) with different concentrations are shown in Fig. 2. From fig.2 indicated that the absorption of the sample increases with increasing EtBr dye doped concentration due to the increase in the number of molecules per unit volume [23]. The absorption coefficients, α, for each sample were determined by the analysis of the optical absorbance spectra and can be obtained from the following relationship [24-28]:

\[ \alpha = \frac{2.303a}{d} \]  

where \( a \) and \( d \) are the absorbance value and thickness of the EtBr dye doped deoxyribonucleic acid sample. Figure 1 shows the presence absorption peak near 486 nm which is the characteristic of DNA. The peak is due to π-π* (where π represents bonding orbitals and π* represents anti-bonding orbitals) transition of the electrons of C=C bond in DNA [29]. At 532 nm and 473 nm the values of \( \alpha \) of the EtBr doped deoxyribonucleic acid samples were calculated and they are given in Tables 1 and 2, respectively.
Fig. 2. UV–visible absorption spectra for EtBr dye doped deoxyribonucleic acid with different concentrations.

3. Z-scan technique

The Z-scan technique is now a standard tool for studying optical nonlinearities in a wide variety of optical materials, because of its high sensitivity and experimental simplicity. In this technique, the EtBr dye-doped deoxyribonucleic acid is translated in the Z-direction along the axis of a focused Gaussian beam, and the far-field intensity is measured as a function of the sample position. While the input power is maintained constant. Scanning measurements were taken in the direction of the Z-axis for both open aperture technique (OA) and closed aperture (CA) technique for the EtBr dye-doped biological polymer using a constant laser power of 4.5mW for solid-state lasers at wavelengths of 473nm and 532nm. To study the effect of wavelength on the Z-scan behavior, two wavelengths 532 nm and 635 nm are chosen for all concentrations samples.

3.1 Nonlinear coefficients

The absorption nonlinear coefficient, $\beta$, can be found using the following relationship [30,31]:

$$\beta = 2\sqrt{2} \frac{\Delta T}{I} L_{\text{eff}}$$

(2)

Since $\Delta T$ is the difference between peck and bottom, $L_{\text{eff}}$ is the effective length and is given by the following relationship [32,33]:

$$L_{\text{eff}} = (1 - \exp(-\alpha L))/\alpha$$

(3)

The amount of the quantity can be defined as the difference between the caliber of the top and bottom of the applied intensity for this type of scanning technique in the direction of the Z-axis. This difference is given by the following relationship [34]:

$$\Delta T_{\text{p-b}} = 0.406(1 - S)^{0.21}[\Delta \phi]$$

(4)

$\Delta \phi$, it represents the phase difference on the axis, and the phase difference on the axis is related to the nonlinear refractive index with the following relationship [35,36]:

$$\Delta \phi = k n^2 I L_{\text{eff}}$$

(5)

As $k$ the wave vector is represented and is given by the following relationship $k = 2\pi / \lambda$ [37], where $\lambda$ it represents the used of laser wavelength and $I_{\text{c}}$ is the intensity value at the focus ($Z = 0$) and is given by the equation [38,39]:

$$I_{\text{c}} = 2P/\pi \omega^2$$

(6)
Therefore, the nonlinear refractive index can be calculated by applying the following equation [40]:

\[ n_2 = \frac{\Delta \phi \lambda}{2\alpha L_{\text{eff}}} \]  

(7)

The change in refractive index is given by the following relationship [41,42]:

\[ n = n_2 I_X \]  

(8)

The linear transmittance of the detector aperture S were equal to 0.61 and 0.73 for the 473 nm and 532 nm wavelength laser, respectively, and in general S is given by [43].

\[ S = 1 - \exp\left(-r_a^2 / \omega_0^2\right) \]  

(9)

Where \( \omega_0 \) is the beam deflection at the detector aperture and it was calculated using the following equation [44]:

\[ \omega_0^2 = \omega^2 \left(1 + \left(\frac{Z}{Z_s}\right)^2\right) \]  

(10)

where \( Z_s \) represent the diffraction length of the beam \( Z_s \) was equal to 3.27 mm, and in general \( Z_s \) is given by the following equation [45]:

\[ Z_s = \frac{\pi \omega_0^2}{\lambda} \]  

(11)

\( Z_s \) represent the distance between the detector aperture and the focal point, \( \omega_0 \) represent the radius of the beam waist at the focus (\( Z = 0 \)), given by the following relationship [46,47]:

\[ \omega_0 = 1.22 f \lambda / D \]  

(12)

where \( f \) is the focal length of the used lens where \( f = +50 \) mm, \( D \) is defined as the diameter of the laser beam at the detector.

3.2. Z-scan technique at 532nm

The open aperture Z-scanning technique was applied on the dye-doped biological polymer (DNA) using a solid-state laser of wavelength 532 nm at the power of 4.5 mW with concentrations (1, 2, 3 and 4 mM) and the measurements were obtained are shown in figure 3.

![Fig. 3: Open Z-Scan technique.](image1)

![Fig. 4: Normalized pure Z-Scan technique.](image2)
The Z-Scan technique of the closed aperture was applied using a laser of wavelength 532nm at a power of 4.5 mW, the measurements obtained were shown in Figure 4, which shows the relationship between the intensity and Z-direction. The Nonlinear optical coefficients values are listed in Table 1.

### Table 1: Nonlinear optical parameters and absorption coefficients at 532nm.

| Co.(mM) | $\beta \times 10^{-3} \text{(cm/W)}$ | $\Delta n \times 10^{-3}$ | $n_2 \times 10^{-7} \text{(cm}^2\text{/W)}$ | $\alpha \text{(cm}^{-1})$ |
|---------|----------------------------------|-------------------------|---------------------------------|------------------|
| 1       | 2.765                            | 0.312                   | 7.965                           | 10.83            |
| 2       | 6.940                            | 0.411                   | 10.499                          | 13.67            |
| 3       | 13.288                           | 0.753                   | 19.218                          | 17.80            |
| 4       | 27.089                           | 1.094                   | 27.936                          | 24.19            |

It has been shown from Table 1 that the values of nonlinear optical parameters of the EtBr dye-doped polymer increase with increasing concentration. To illustrate this effect more accurately and clearly, Figure 5 shows the nonlinear absorption coefficient and nonlinear refractive index as a function of concentration, as it can be seen from the curve that the relationship between them is linear, that is, when the concentration increases, the nonlinear absorption coefficient and nonlinear refractive index is a linear function.

![Figure 5](image1.png)

**Fig. 5:** $n_2$ and $\beta$ as a function of concentration of dye doped biological polymer.

### 3.3. Z-Scan technique at 473nm

With the same power of 4.5mW for the 473nm wavelength laser, this technique was applied, and the measurement was obtained. Figure 6 shows the relationship between the normalized transmitted of the sample as a function of the location when the aperture is opened.

![Figure 6](image2.png)

**Fig. 6:** Open Z-Scan technique.

![Figure 7](image3.png)

**Fig. 7:** Normalized pure Z-Scan technique

Scanning measurements were taken with the direction of the Z-axis of dye doped biological polymer (DNA) at the power of 4.5 mW using a solid-state laser with a wavelength of 473 nm. The measurements obtained were
shown in figure 7, which shows the relationship between the normalized transmitted and the sample location, for pure Z-scan data. Table 2 shows Z-scan nonlinear coefficients values of the dye-doped biological polymer at the wavelength of 473nm.

Table 2: Nonlinear optical parameters and absorption coefficients at 473nm.

| $C_0$(mM) | $\beta \times 10^{-3}$ (cm/W) | $\Delta n \times 10^{-3}$ | $n_2 \times 10^{-2}$ (cm$^2$/W) | $\alpha$(cm$^{-1}$) |
|---------|-----------------|-----------------|-----------------|-----------------|
| 1       | 5.45            | 0.49            | 8.49            | 24.08           |
| 2       | 8.05            | 0.70            | 11.77           | 28.99           |
| 3       | 33.42           | 1.34            | 22.54           | 35.56           |
| 4       | 67.45           | 2.51            | 42.36           | 45.51           |

The results shown in Table 1 and 2 have shown that the nonlinear parameter values of the EtBr dye-doped biopolymer DNA, which are the nonlinear absorption coefficient, the nonlinear refractive index and the phase difference, increase with increasing concentration at the wavelength of 473nm more than at the wavelength of 532 nm. Figure 8 shows the nonlinear absorption coefficients and the nonlinear refractive indices at the wavelength of 473 nm as a function of sample concentration.

![Fig. 8: $n_2$ and $\beta$ as a function of concentration of dye doped biological polymer.](image_url)

4. Spot size behavior.

During the Z-scan technique, the scanning process is along the Z-axis in positive (+Z) and negative (-Z) directions. Therefore, the sample is in different locations for the focus and thus, the spot size is constantly changing according to the location. We deliberately studied this continuous change in shape, where we dispensed with the detector device by replacing it with a sensitive screen. The spot size was studied as a function of the Z-scan location of the dye-doped DNA polymer at a concentration of 3 mM. The spots size of the laser beam (the penetrating beam) were taken on a screen placed at a distance from the sample at the location in which the intensity appears (vertex) along the Z-axis, and the location in which the intensity appears (bottom) by a digital camera. The Z-scan technique was performed using a linearly TEM$_{00}$ Gaussian beam of solid-state CW diode laser at 4.5 mW ($\lambda$=473 nm). Different spot size shape for the selected scanning locations has been appearing on the sensitive screen. Fig. 9 shows the characteristics of the obtained spot size.
Fig. 9: change in the laser spot size along Z-position.

Fig. 9 displays the pure z-scan results of EtBr dye-doped DNA polymer. While the samples move along the Z-axis in the measurements, the incident fluence increases from a lower value to its maximum value. At the Z-position 'far' away from the focal point, this means low input fluence. The samples exhibit linear optical behavior. The size of the spot is as small as possible at the valley, and if the intensity is very high because the effect of non-concentration contributes to the nonlinear heat as a result of the absorption of the laser beam according to the absorption of the sample, this effect has the maximum permeability as a result of saturation at high intensity[48-50].

5. Conclusions:

In conclusion, we studied the third-order nonlinear optical response of Ethidium bromide doped deoxyribonucleic acid (DNA) by performing both open-aperture and closed-aperture Z-scan technique with a TEM00, Gaussian solid state CW laser at 532 nm and 473 nm. The third-order nonlinear refractive index was as large as $42.36 \times 10^{-7} \text{cm}^2/\text{W}$ has been achieved. The values of $n_2$ and $\beta$ obtained at 532 nm are the same order as those obtained at 473 nm. The nonlinear refractive index had a negative sign, which was due to self-defocusing. The magnitude of $n_2$ with the negative sign and $\beta$ were in the order of $10^{-7} \text{cm}^2/\text{W}$ and $10^{-3} \text{cm}/\text{W}$, respectively. They were dependent on the concentrations of Ethidium bromide dye. The doping of the DNA polymer by the EtBr dye worked to improve the ability of the material to show high optical properties of nonlinearity, as the denaturation worked on doped the DNA polymer and the binding was high with the dye through the bases of Adenine (A) and Guanine (G) as well as the association with the three bases of bermdene. Figure 9 shows the comparison between pure and Ethidium bromide doped DNA and how the DNA bases are related when doped.

Fig. 10: Comparison of pure and Ethidium bromide dye doped DNA.
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