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PHYSICS

A Home –Made of Static Laser Scattering Experiment for Dopamine Investigations

R. Ghazy[1]*, F. Shehata[1], F. El-Mekawey[1], N. Sayed[2]

1 Laser Lab. Physics Dept.; Faculty of Science; Tanta Univ.; Tanta; Egypt.

2 Bio-physics Dept.; Faculty of Sciences; Cairo Univ.; Cairo; Egypt.

*Corresponding author E-mail addresses:

riad.ghazi@science.tanta.edu.eg، riyadghazy@yahoo.com.

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KEY WORDS

Dopamine, Dynamic laser scattering, Static laser scattering.

ABSTRACT

A Self home-built of static laser scattering experiment was made to investigate the scattering effects of various polymer solutions at various concentrations to recognize the scattering parameters. The scattering parameters of a selected material such as molecular weight, the radius of gyration, and the second virial coefficient were determined at room temperature. The investigated material that we have used is Dopamine, which is known as the happiness hormone. With the aid of refractive index measurements at various concentrations, calculation of its index increment and the symmetrical behavior of the scattering phenomena with regard to the angle of the scattering, the scattering parameters are investigated. In this study we have used what is called Zimm Plot method.

By using a diode laser as a light source for light scattering, a diode laser was executed at a wavelength of 650nm.The values that were assessed of the refractive index increment, the molecular weight, the second virial coefficient and the radius of gyration were observed to be 7.33ml/g, 223.555 g/mol, 0.0495651mol.ml/g^2, and 2.5836nm, sequently.

1. Introduction

Dopamine is a naturally occurring monoamine that has two hydroxyl substituents on the benzene ring as well as an aminoethyl group linked to the substituted ring, in addition, it is a type of neurotransmitter relating to a family of "catecholamines," that also comprises numerous relevant neurotransmitters [1].Dopamine, norepinephrine, as well as epinephrine are the three main neurotransmitters.

Dopamine is a hormone that plays a significant role in the brain and body. It acts as a chemical messenger between neurons, once your brain identifies activity with pleasure, motivation, or excitement, and reward it releases dopamine. Most diseases of the nervous system are connected with dysfunctions of dopamine as Parkinson's disease, depression and dopamine transporter deficiency Syndrome [2]. The biosynthesis process of dopamine through tyrosine hydroxylase converts tyrosine to L-DOPA, while aromatic L-amino acid decarboxylase converts tyrosine to dopamine, so the pathway is Tyrosine → L-DOPA → Dopamine [3].

D_1 and D_2 are the dopamine receptors most frequently expressed in the brain (D1 being the highest), and both are rarely co-expressed into the same cells. The brain activities that regulate mood, sleep, memory, education, focus, and motor control all have an impact on dopamine levels in the body. Certain medical conditions such as depression or Parkinson's disease may be related to a dopamine deficit. A drop in the body's dopamine or a receptor problem in the brain can result in a dopamine deficiency [4].

The majority of mental health disorders and problems, however, they are frequently related to too much or too little dopamine in various parts of the brain. Here are some examples:-

1-Parkinson's disease [5] is a neurological disorder in which people have problems controlling voluntary muscle movement, dopamine levels are significantly lower in Parkinson's disease patients.

Parkinson's disease is the most frequent type of neurodegenerative disease which shown by older patients because the gradual loss of dopamine cells allowing the disease to manifest. Due to loss of dopaminergic neurons, there is a considerable decrease in circulating DA.

2- Abuse of drug and addiction like cocaine causes DA levels increase through blocking the dopamine carrier activity. Even though, drugs reduce the ability of your body to naturally obtain dopamine [6].Dopamine as a hormone is very crucial in the human body so the physicochemical characteristics of dopamine are valuable to characterize.

Light scattering techniques are divided into two types: static and dynamic light scattering [7,8] .Both techniques can be used to characterize macromolecules and a wide spectrum of particles in liquids quantitatively, when the incident light's frequency is the same as the scattered light, it is known as static (elastic) scattering [8-10].Unlike dynamic light scattering (inelastic) that occurs when the incident light's frequency is variant from the scattered light’s [9,10]. So, Multi-angle static laser Light scattering is an efficient method to characterize the structure of polymers, nanoparticles as well [9]. In this research, laser scattering is used to determine the molecular weight, the second virial coefficient and the radius of gyration for dopamine.

Light scattering can be defined as the passage of laser light throughout the colloidal solution, after which the particles absorb incident light and vibrate as the incident light frequency. As a result of this vibration, the particles re-emit the light throughout all directions at the same frequency [11]. For big molecules, the molecular weight, the radius of gyration, and the second virial coefficient are the basic characteristics provided by measuring light scattering in a dilute polymer solution [12, 13]. Debye's equation confirmed that the scattering medium's molecular weight is connected to the intensity of scattered light and its angular distribution [14].The angular distribution of scattered light provides vital data about the physical qualities and scattered medium parameters [15,16]. The Debye equation demonstrates the necessity of monitoring refractive index increment when evaluating scattering parameter.

As a result, considerable effort has been expended in enhancing the device that uses the laser beam because the beam is monochromatic, it has a high degree of coherence as well, and measurements of scattered light have been more impressive.

In this paper, we designed a laser scattering apparatus and evaluated the scattered laser light's angular distribution. We calculated the molecular weight M_w, the radius of gyration R_G, and the second virial coefficient A_2 using these data. Dopamine was chosen, with toluene serving as our system standard.

Calculations were achieved using a multi-angle light static laser scattering experiment, the light scattered at various angles from a clear solution is depicted in Fig. (1), of dopamine. The rectangular cell was filled with different concentrations of dopamine and must be stable to be examined with a photomultiplier tube mounted on a rotary table, while the rotary table of the spectrometer has a photomultiplier tube that can be rotated around the sample at various angles at 40° to 140°scattering intensity measurement. A toluene angle of 90^o was used to calibrate the measurements. [17]. A diode laser with a wavelength of 650 nm was applied as a light source. By measuring the intensity we can calculate scattering parameters as molecular weight (M_W), second viral coefficient (A_2) and the square radius of gyration $<R_G^2>$ are computed using the Zimm plot method. Abbe refractometer was employed in order to assess the refractive index *n* and the refractive index increment dn/dc for different concentrations.

2. Materials

2.1. Sample Preparation

0.01 gram of dopamine hydrochlorides was dissolved in 40 ml of purified water, this is the stock which used to prepare several concentrations (0.000025, 0.0005, 0.00075, 0.001 and 0.00125 g/ml). Dopamine hydrochlorides were obtained from (Sigma Aldrich).

3. Results and discussion.

3.1. Verifying of refractive index increment.

To measure refractive index increment of dopamine that dissolved in purified water as a function of concentrations (0.000025, 0.00005, 0.000075, 0.0001 and 0.000125) by using Abbe refractometer. At first, the refractive index (n) is measured then the sample was adjusted for the measuring prism and measure the reference refractive index (n_o) , then the refractive index of various concentrations of dopamine are measured. The relationship between (n) refractive indices and (C) is graphed as shown in Fig. (2). after that the relation between (Δn) (the difference between the measured refractive index and reference refractive index) and(C) concentration as shown in Fig. (3). by executing increments on various dopamine concentrations, the concentration dependence was found. The dopamine's refractive index increment dn/dc is computed as illustrated in Fig (4), as well as the quantity of Δn/c versus c is graphed in order to compute the value of $\lim_{c\to 0}\frac{\Delta n}{c}$.

3.2 Static (elastic) light scattering

The Rayleigh scattering theory is an approach that describes how light scatters by Rayleigh so if the particle diameter is less than λ /20, the study has the following objectives: - Light scattering intensity, refractive index increment, the molecular weight ,the second virial coefficient, in addition to, the radius of gyration are all connected by the Debye equation [12].For small molecules (λ/20) appear to an incident beam as a point and scatter light symmetrically throughout all directions. Light waves emitted by the molecule's various scattering centers will have a relatively insignificant phase difference. The intensity of the light scattering can be calculated regardless of the scattering angle in this case [9].

$$\frac{KC}{R_\theta} = \frac{1}{M_w} + 2A_2C + 3A_3C^3 \qquad (1)$$

Where;

A_2 : The second virial coefficients, A_3: The third virial coefficients, M_w Molecular weight, C: Concentration of Solute, R_θ : Rayleigh ratio that can be defined as shown is Eq. (2) [18]. The Rayleigh ratio is a measurement of the sample's absolute scattering that is unaffected by experimental parameters like the distance between the detector and the sample (r) or volume of scattering V [19].The Rayleigh ratio's physical concept is the incident beam's dissipation caused by intensity loss after passing through a medium[18].

$$R_\theta = \frac{I_\theta}{I_0}\frac{r^2}{V} \qquad \textbf{(2)}$$

Where;

I_θ : Scattered light intensity, I_0 : incident light intensity,

r : distance between the photomultiplier tube shutter as well as the sample; and V: the volume of scattering .

The physical parameters of dopamine were determined by measuring the angular distribution of scattered light intensity using an experimental static scattering technique, as depicted in Fig. (4).

The basic equation explaining light from diluted solutions for non-uniform big macromolecules is commonly expressed in the form at finite concentrations. The molecule's size and the second virial coefficient aren't absolute values; however, this is dependent on the solvent. As a result, good solvents have a larger gyration radius than bad solvents [20].

The refractive index increment dn/dc is included in the optical constant K in that can be calculated from Eq. (4)

$$K=\frac{2\pi^2 n_o^2}{\lambda^4 N_A}\left(\frac{dn}{dc}\right)(1+\cos^2\theta) \qquad (4)$$

Where N_A stands for the number of Avogadro, λ the scattered light's wavelength, n_o represents the solvent's refractive index; and θ is the angle of scattering.

For dopamine dissolved in distilled water at various concentrations of 0.000025, 0.0005, 0.00075, 0.001 and 0.00125 g/ml, the angular distribution (I / I_0) is shown in Fig. (5). The intensity of scattered light was measured at angles ranging from 40^o to 140^o. The Rayleigh equation can be used to describe the observed behavior of the scattered light ratio I/I_0 as a function of scattering angle, which is given by Eq (5) [18].

$$\frac{I}{I_o}=\frac{16\,\pi^4\alpha^2}{\lambda^4 r^2}(1+\cos^2\theta) \qquad (5)$$

Where; α known as the polarizability of a molecule. The acquired results are graphically represented as kc/R_θ vs. c as well as kc/R_θ vs. $\sin^2(\theta/2)$ as shown in Fig. (6) and Fig. (7) respectively. Observations produce a sequence of straight lines, in either instance kc/R_θ vs. c at constant θ as well as therefore kc/R_θ vs $\sin^2(\theta/2)$ at constant c as shown in Fig.(6) and Fig.(7) respectively. These lines might be extrapolated in order to allow the estimation of the kc/R_θ at c equals zero, as well as at θ equals zero, so in Eq. (3) The phrase $2A_2c= 0$ is in the first instance, the phrase $\left(\frac{16\pi^2}{3\lambda^2}\right) <R_g^2> \sin^2\left(\frac{\theta}{2}\right) = 0$ is in the other instance .

At C= 0, we get:

$$\frac{KC}{R_\theta}=\frac{1}{M_w}\left[1+\frac{16\pi^2}{3\lambda^2}<R_g^2>\sin^2\left(\frac{\theta}{2}\right)\right]. \qquad (6)$$

At $\sin^2\left(\frac{\theta}{2}\right) = 0$, we get:

$$\frac{KC}{R_\theta}=\frac{1}{M_w}+2A_2C \qquad (7)$$

Plotting the extrapolation values for $lim_{c\to 0}\frac{KC}{R_\theta}$ for $\theta_1, \theta_2, \theta_3, \theta_4, \theta_5$ and θ_6 versus $\sin^2\left(\frac{\theta}{2}\right)$, Eq. (6) yields a line with assumptions related to those values .Similarly, the $\frac{KC}{R_\theta}$ numbers for the various concentration c_1, c_2, c_3 and c_4 for $\sin^2\left(\frac{\theta}{2}\right)$ can be acquired through extrapolation .If these value extrapolates are drawn in accordance with concentration c, similarly, we get the straight line that corresponds to Eq. (7). The intercept at the ordinate for both straight lines is $1/M_w$ [21, 22] as shown in Fig.(8) and Fig.(9) respectively . The value of molecular weight can be computed the slope of a straight line at C = 0 with the following formula:

$$\frac{1}{M_w}\left(\frac{16\pi^2}{3\lambda^2}\right)<R_g^2>\sin^2\left(\frac{\theta}{2}\right).$$

And through this we obtain the $<R_g^2>$ (the radius of gyration). By plotting the relation between kc/R_θ and C since $\sin^2(\frac{\theta}{2})$ = 0, the line's slope is exactly equivalent to $2A_2$ (double of the second virial coefficient).

3.3 Zimm plot method.

We can calculate the radius of gyration, molecular weight, as well as the second virial coefficient A_2 utilizing Zimm's graphing method from light scattering measurements at varied angles and concentrations. Generally, in a single diagram displaying KC/R_θ vs $\sin^2(\frac{\theta}{2})$ +KC. Selecting the k value to make the concentration term similar to $\sin^2(\frac{\theta}{2})$.

The constant-angle line to c=0 should be extrapolated before the line of constant concentration is extrapolated to θ=0 due to the lower angles' uncertainty. It It generates a graph with grid lines, which, with the same intercept, must fulfill two lines of which KC/R_θ to c= 0 as well as KC/R_θ to $\sin^2(\frac{\theta}{2})$ =0 to extrapolate and which is the validity requirement of Eq. (3).The intercept reciprocal [KC/R_θ] when C = 0, θ = 0 is equals the weight of the molecular average .

The values measured by the (I/I_o) laser light intensities are displayed in Fig. (5) to indicate the angular distribution in various scattering angles respectively.

Different values were determined according to concentration, for KC/R_θ all at a constant observer angles $\theta_1=40^o$, $\theta_2=50^o$, $\theta_3=60^0$, $\theta_4=70^o$, $\theta_5=80^o$, and $\theta_6=90^o$.

Similarly, in the same way KC/R_θ values are displayed in relation to $\sin^2(\frac{\theta}{2})$, as illustrated in Fig. (6) for the constant concentrations of c_1= 2.5e-^{5,} c_2= 5e^{-5}, c_3= 7.5e-^{5} c_4=1.0000e-^{4}, and c_5=1.2500e^{-4} g/ml.

Whenever these extrapolation results are graphed as concentration, c, vs for which c=0, the data relating to these values are shown in Fig. (7), which demonstrates that a straight line is obtained according to Eq. (9).Similarly, the KC/R_θcalculated values at $\sin^2(\frac{\theta}{2})$ = 0 can be determined by extrapolating the KC/R_θ calculated values versus the concentration as illustrated in Fig. (8).

Straight lines relating to equations (9, 10) were produced from the Fig. (9).The intercept on the ordinate for both straight lines is 1/Mw. The slope of the straight line can be used to calculate the value of $<R_g^2>$ from the line at c = 0. There are 30 data points for a Zimm plot with five distinct concentrations and six scattering angles, and a zero concentration line would be six points and a zero scattering angle would be four points so Fig. (10) displays a plot of Zimm for dopamine, with the results in Table (1).

Table (1): the refractive index n, the refractive index increment dn/dc, the molecular weight Mw, the radius of gyration Rg, and the second virial coefficient A_2 of dopamine solutions that determined at room temperature.

Concentrations (g/ml)	dn /dc (ml/g)	M_w (g/ml)	R_g (nm)	A_2 (mol.ml/g^2)
(2.5e-^{5,} 5e^{-5}, 7.5e-^{5} 1.0000e-^{4}, and 1.2500e-^{4}	7.33	223.555	2.5836	0.0495651

4. Conclusion

The molecular weight is determined using static laser light scattering for any matter that is unknown for us, also we make use of angular distribution for various angles from (40 to 140) to know the scattered intensity. We measured also radius of gyration, the second virial coefficient by using Zimm plot so the molecular weight, the second virial coefficient as well as the radius of gyration were found to be 223.555 g/mol, 0.0495651mol.ml/g^2, and 2.5836nm, respectively.

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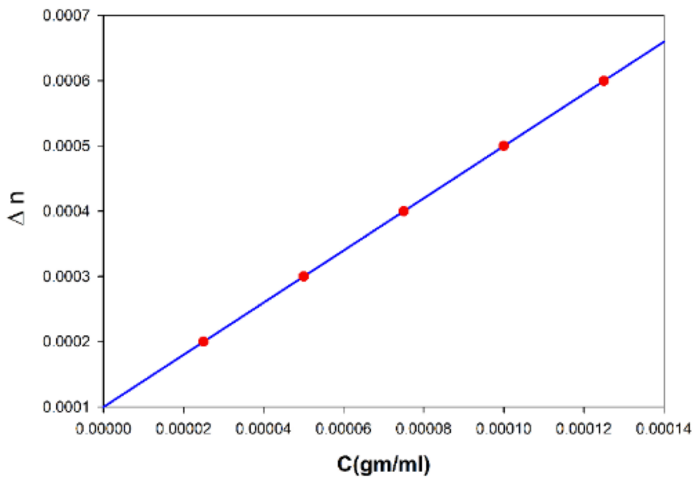

Fig. (1): The relation between the refractive index n of dopamine at different concentrations c.

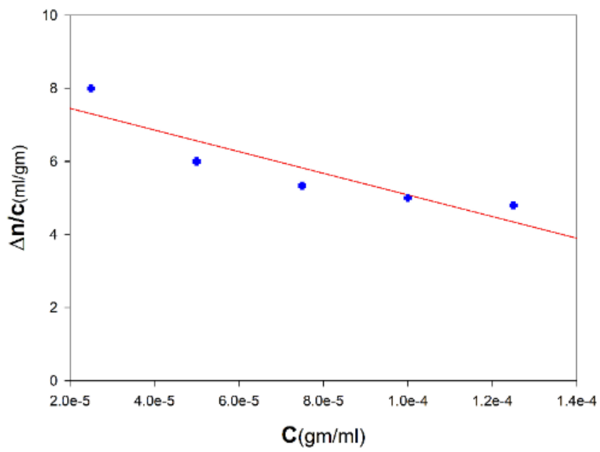

Fig. (2): The relationship between the differences in refractive index n as a function of concentration c.

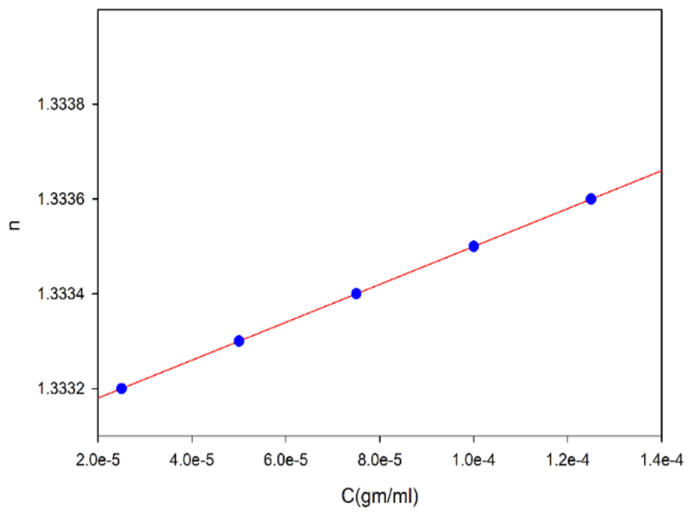

Fig. (3): The dependence of Δn/c on the various dopamine concentrations c.

4. System set-up

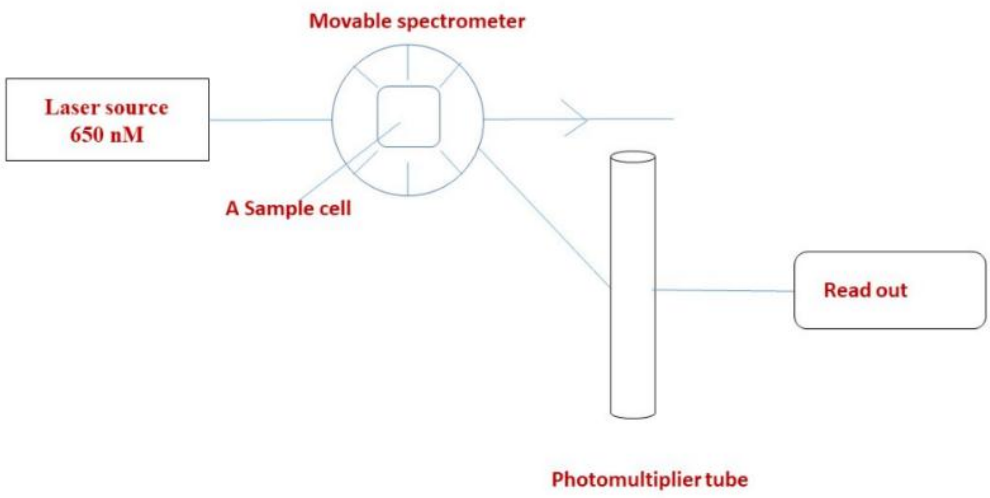

Fig. (4). A home –Made of Static Laser Scattering Experiment.

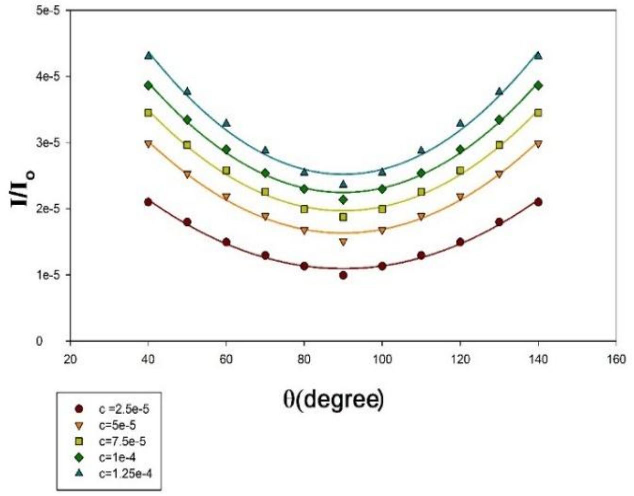

Fig. (5): The angular distribution of scattered light intensity of dopamine.

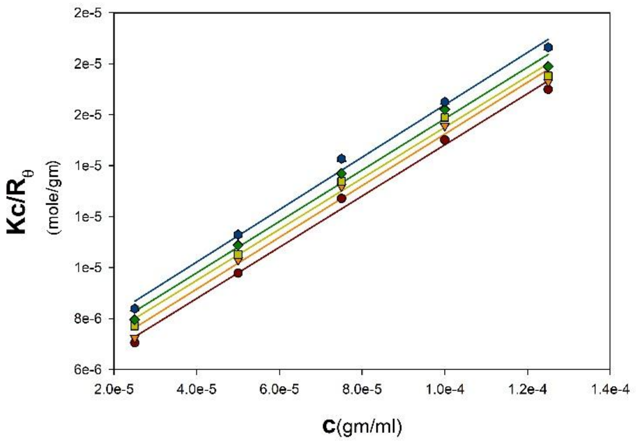

Fig. (6): Kc/ $R_θ$ for different angles versus concentration.

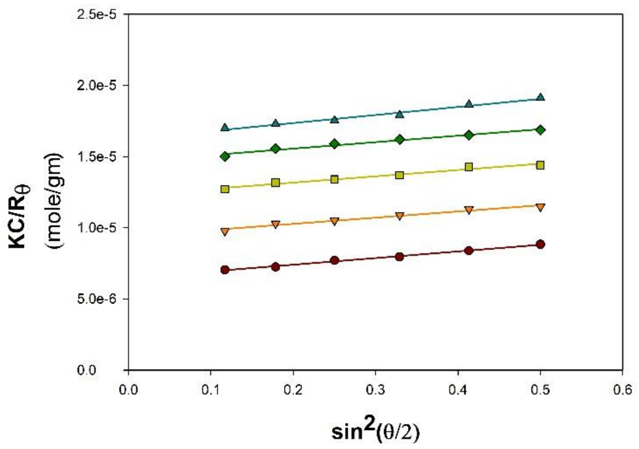

Fig. (7): Kc/Rθ for different concentrations versus $\sin^2(\theta/2)$

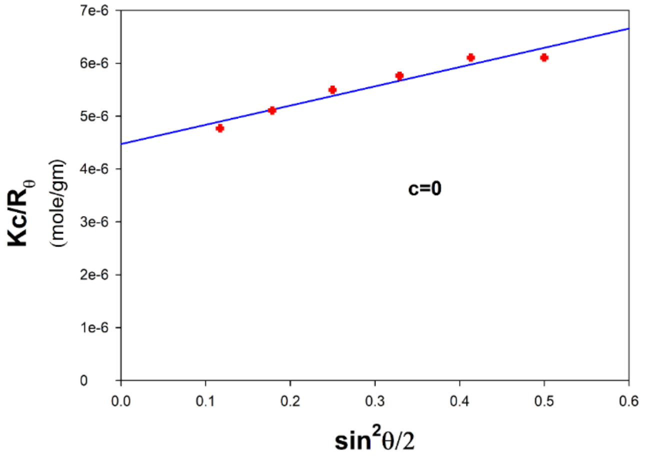

Fig. (8): The dependence of (Kc/R_θ) on $\sin^2(\theta/2)$ at zero concentration of dopamine.

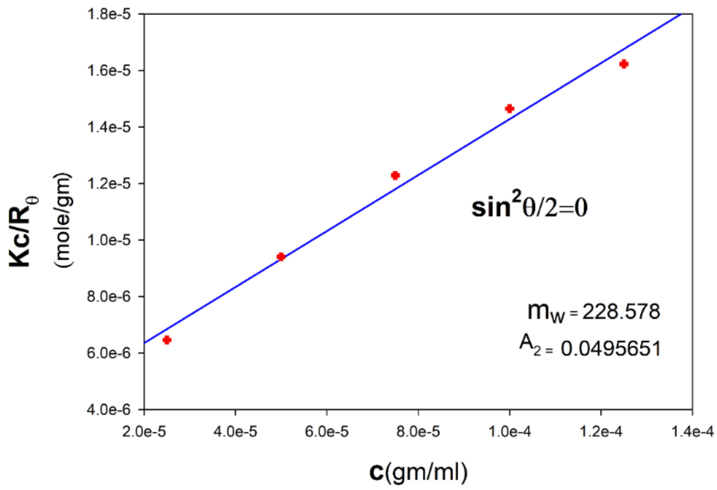

Fig. (9): The dependence of (Kc/Rθ) on concentration at $\sin^2(\theta/2) = 0$.

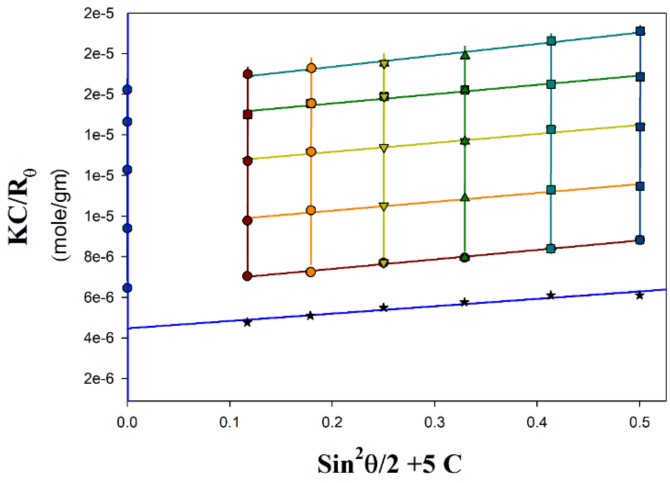

Fig. (10): The zimm plot for dopamine at different concentrations and scattering angles.

بناء تجربه الاستطاره الساكنه لدراسه هرمون الدوبامين

ا.د/ رياض غازي[١] ، فاطمه شحاته[١] ، ا.د / فاروق المكاوي[١]، د/ نهي سيد[٢]
١- كلية العلوم - جامعه طنطا- مصر
٢- كلية العلوم - جامعه القاهره , مصر

تم انشاء تجربه الاستطاره الساكنه باستخدام شعاع الليزر داخل المعمل حيث تم دراسه ظاهره الاستطاره الساكنه لمحاليل هرمون الدوبامين محل الدراسه وذ لك باعداد تركيزات مختلفه لذات الماده.

وتم تعيين معاملات الاستطاره وهي الوزن الجزيئي ونصف قطر دوران الجزيئى ومعامل التخمين من الدرجه التانيه وتم اجراء هذه الدراسه عند درجه حراره الغرفه للماده محل الدراسه .

وقد تم اختيار هذه الماده لدراسه مدي فاعليه الكشف عن هرمون الدوبامين والذي يعرف بهرمون السعاده وذلك بمساعده تعيين معامل الانكسار ومعدل زيادته كداله في التركيز .

اوتممت هذه الدراسه بقياس شده استطاره شعاع الليزر كداله في زاويه الاستطاره وقد تم حساب معاملات الاستطاره باستخدام طريقه زيم (Zimm plot).

وذلك باستخدام شعاع الليزر من النوع دايود بطول موجي٦٥٠ نانومتر عند تركيزات مختلفه للماده محل الدراسه.