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Low Level Laser Therapy For Hyperoxia-Induced Retinal Protein Deformations Evaluated By FTIR Study

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THE aim of the present study was to evaluate the efficacy of low-level light emitted diode laser (LED) therapy on retinal protein of albino rats exposed to high concentration of oxygen (O_2). Fifty Wistar albino rats were divided into three groups as follows: (a) control group (n=10 rats) without neither hyperoxia exposure nor laser treatment, (b) hyperoxia group (n=20 rats) received daily high concentration of O_2 (90%), for one week (n=10 rats) and two weeks (n=10 rats) (c) LED laser treated group: hyperoxia group (n=20 rats) as the previous group was treated with a 670 nm light emitted diode laser two sessions a week. Rats for experimental groups were sacrificed after one week and two weeks (n= 10 eyes rats each), Fourier transform infrared spectrum analysis was applied.

Confirmed that retinal protein analysis revealed structural deformation that appeared after hyperoxia in NH–OH and fingerprint regions. $_{str}O–H_{sym}$ band shifted from 3265±1 cm^{-1} to 3284±1 cm^{-1} ($p<0.01$) and $O\text{-}H_{str}$ band width was increased from 118±1 to 150±2 ($p<0.01$). CH3 deformation and COO_{assy} bands are mostly affected in fingerprint region. Wavenumber of the first one was moved from 1294 ٢± cm^{-1} to 1327±١ cm^{-1} ($p<0.01$) where the second band was missed. After treatment with LED laser, amide I frequency may be enhanced in both one week and two weeks with hyperoxia groups. Bands of amide II, COO_{sym}, CH_3 Deformation and PO_2 asymmetric have non-significant change in their vibrational frequencies ($p>0.05$), Using low level LED laser therapy showed a clear improvement in the retinal protein structure due to its ability to treat and/or reduce the progression of retinopathy and conditions involving cell death. which illustrated by Fourier transform infrared analysis.

Keywords: Hyperoxia, FTIR spectroscopy, Low level laser, Retinal protein

Introduction

Hyperoxia arises when tissues and organs are exposed to an excess supply of oxygen (O_2) with high partial pressure, high concentrations and long durations. In medicine, it alludes to the overabundance oxygen in lungs or other body tissues, which can be brought about by breathing air or oxygen at pressures more prominent than typical atmospheric pressure (21%) (Mach et al., 2011). This sort of hyperoxia can prompt oxygen lethality. In intensive care unit ICU patients, hyperoxia was correlated with mortality (Helmerhorst et al., 2017). As well as, the increased death rate has been observed in patients with traumatic brain injury and worse functional outcomes related to hyperoxia (Rincon et al., 2014). In the retina, early researches in neonatal, adult rabbits, and adult mice proved that exposure to hyperoxia can produce serious impairment to photoreceptors. The most spotted morbid effects in the hyperoxia-induced mice are photoreceptor and endothelial cell death, increased inflammation, oxidative stress production, breakdown of the blood-retina barrier (BRB) and vision loss (Albarracin et al., 2013). A new method has emerged recently to treat many common diseases, such as treatment of infected, ischemic, diabetic wounds and other soft tissue injuries in humans and animals. This method can be considered with no hazard effects on the living system, which is the use of near infrared (NIR) via light emitted diode (LED) laser therapy with wavelength range (600-1000) nm (Eells et al., 2004). This regimen is characterized by many useful features such as wound healing, limiting tissue damage, decreasing

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inflammation and inhibition of tissue pathology (Abdelkawi et al., 2018). In the past 10 years, there has been obvious interest to use 670 nm red light for retinal injuries treatment. Recently, it was emphasized the effectiveness of using 670 nm red light to moderate the damaging effects of methanol intoxication in rat retinas (Albarracin et al., 2013) . This study aimed to explore the efficacy of LED laser therapy in the treatment of hyperoxia retinal degenerations assessed by Fourier transform infrared spectroscopy (FTIR).

Materials and Methods

Experimental animals

Fifty female's Westar albino rats were involved the rat's ages were ranged from 8 to 12 weeks; their weight was 200±20 g. The rats were maintained in a standard 12 hr. light- dark cycle with free access to water and balanced diet at a temperature of 25 ± 2°C, 50% humidity. Examination for rats' eyes by slit lamp bio microscope before exposure to hyperoxia revealed no signs of edema or intraocular inflammation.

Hyperoxia exposure

Experimental animals divided into two main groups (a) Ten rats were served as control group which did not receive any treatment (n=10 rat). (b) Fourty rats (n=40 rat) were kept in glass chamber and exposed to continuous flow of 90% O_2 concentrations (hyperoxia group) for 20 hr/ day for periods of one and two weeks, The hyperoxia group were divided into two subgroups (1b) hyperoxia group (n=20 rat) did not receive any treatment and (2b) other hyperoxia group exposed to low level LED laser (n=20 rat).

Low level LED laser protocol

Rats were anesthetized by intramuscular injection with a mixture of 45 mg/ kg ketamine hydrochloride (Ketalar ®, Sandoz Canada Inc.) and 21 mg/ kg pentobarbital sodium (Rematal® Sodium Taj Pharmaceuticals Limited, India). Low level diode laser (670 nm) was generated from continuous wave (CW) diode-pumped solid-state laser (Cobolt, DPSSL-DRIVER II, China) and calibrated to deliver a beam power of 50 mW/ cm^2, 90 sec (2 sessions / one week) using a laser prop and convex lens for beam focusing. The total radiant exposure energy was thus 50x90x2=9000 mJ/ cm^2 or 9 J/cm^2 for each eye.

Extraction of retinal protein

Rats were decapitated after one week and two weeks; the eyes were enucleated and the retinas were carefully removed from the posterior chamber of the eye. Extraction of retinal layers was carried out and directly prepared for FTIR.

Fourier Transformer Infrared Spectroscopy (FTIR)

The retinal cells were freeze dried and mixed with KBr powder (98mg KBr: 2mg of lyophilized retina) to prepare the KBr disks. FTIR spectra were measured in the range of 4000-1000 cm^{-1} at room temperature using Thermo Nicolet iS5 FTIR spectrometer (Thermo Fisher Scientific Inc, Madison, USA) with effective resolution of 2 cm^{-1}. Hundred sample interferograms were recorded for each spectrum. The spectrometer is operated under a continuous dry nitrogen gas purge to get rid of interference from atmospheric carbon dioxide and water vapor. The data was baseline corrected and smoothed by Savitzky-Golay to remove the noise before Fourier transformation. The obtained group spectrum was normalized and analyzed for the following spectral regions: (i) NH-OH region at 3700-3000 cm^{-1}; (ii) CH stretching region at 3000-2800 cm^{-1}; (iii) fingerprint region at (800-1800) cm^{-1} which includes the amide I band (1800-1600 cm^{-1}). The average of the individual spectrum for each group was obtained using Origin Pro 9 software.

Statistical evaluation

Data was expressed as the mean ± SD (Standard Deviation) and the analysis of variance (ANOVA) procedure was used for comparison between different groups, where a commercially available software package (SPSS-11, for windows) was used and the result was considered significance at $P < 0.05$.

Results

NH-OH region

The IR frequency range at 3000-3800 cm^{-1} corresponding to stretching NH-OH region after one week was shown in Figure (1). The wave numbers and bandwidths of the deconvoluted peaks for control, hyperoxia and LED laser treated group after one week were listed in Table (1). By applying the curve enhancement procedure, the control retina revealed three characteristic bands appeared at 3607 cm^{-1}, 3472 $cm^{-1}$and 3265 cm^{-1} corresponding to $O–H_{str}$, $_{str}O–H$ and $_{str}O–H_{sym}$ respectively (Dovbeshko et al., 2000) . For the hyperoxia group, the band at 3607cm^{-1} attributed to $O–H_{str}$ was shifted to 3592±1cm^{-1} ($p<0.05$) accompanied with obvious increase in bandwidth. The vibrational frequency of $_{str}O–H$ band was decreased to 3460±3 ($p<0.01$) and in bandwidth, $_{str}O–H_{sym}$ band vibrational frequency was moved to 3284±1cm^{-1} ($p<0.01$).

In LED laser treated group, the band corresponded to O–H_{str} at 3609±1cm^{-1} became close in values of frequency and bandwidth to the control group. In addition, the other two bands ($_{str}$O–H and $_{str}$O–H_{sym}) were critically improved and enhanced better than hyperoxia group but still less than control one.

After two weeks of hyperoxia (Figure 2) the detected changes in wave number and/or bandwidth after one week of hyperoxia still persist. In LED laser treated group, the two bands corresponding to OH_{str} and $_{str}$O–H were recovered and became near to the control group (Table 1). Besides, there was significant improvement in the wave number and bandwidth corresponding to $_{str}$O–H_{sym} ($p>0.01$).

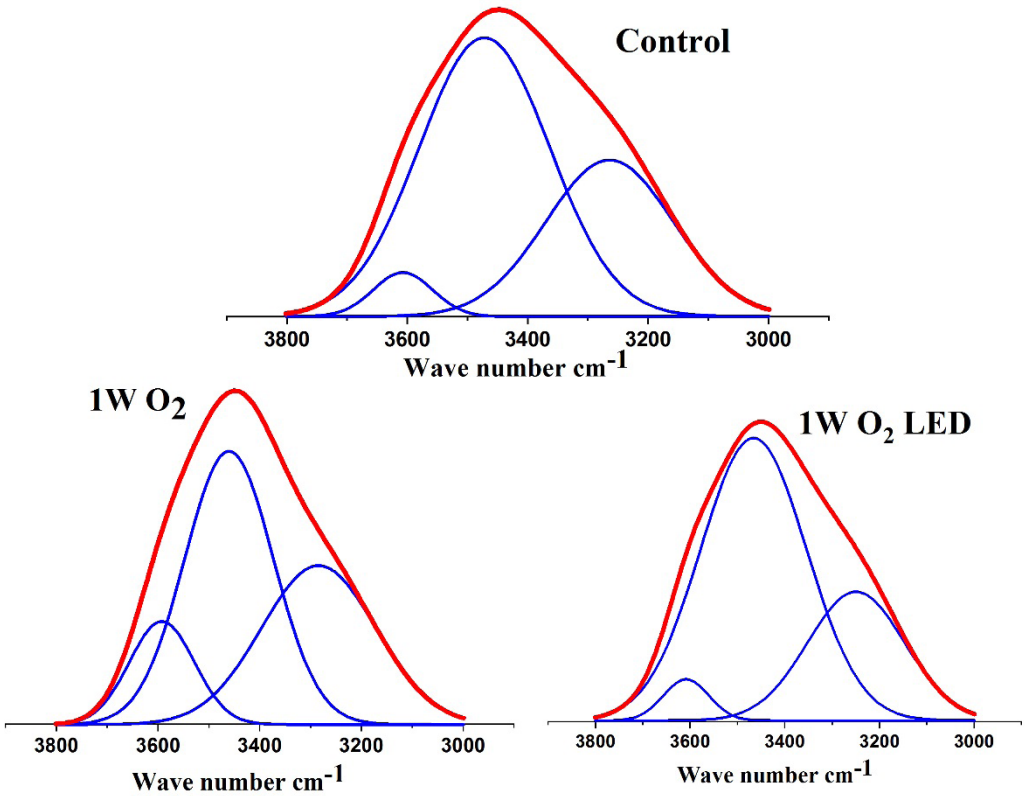

Fig. 1: FTIR spectra of the control retina, after one week of hyperoxia (1W O_2), and treated with LED laser (1W O_2 LED) groups in the NH-OH stretching region (3700- 3000 cm^{-1}).

TABLE 1. FTIR results of the retina for control, hyperoxia and LED laser treated groups. NH-OH region (3000-4000 cm^{-1}) after one week and two weeks, first line indicates wave number and the second line for bandwidth.

Peak	control	1W O_2	2W O_2	1W O_2 LED	2W O_2 LED
O-$_{Hst}$r	3607±1 118±1	3592±1 150±2	3592±1 150±2	3609±1 114±2	3609±1 131±1
$_{str}$O–H	3472±1 254±1	3460±3 205±3	3460±3 206±3	3466±1 265±2	3465±1 259±1
$_{str}$O–H_{sym}	3265±1 250±1	3284±1 265±3	3284±1 264±3	3260±1 241±1	3263±1 238±1

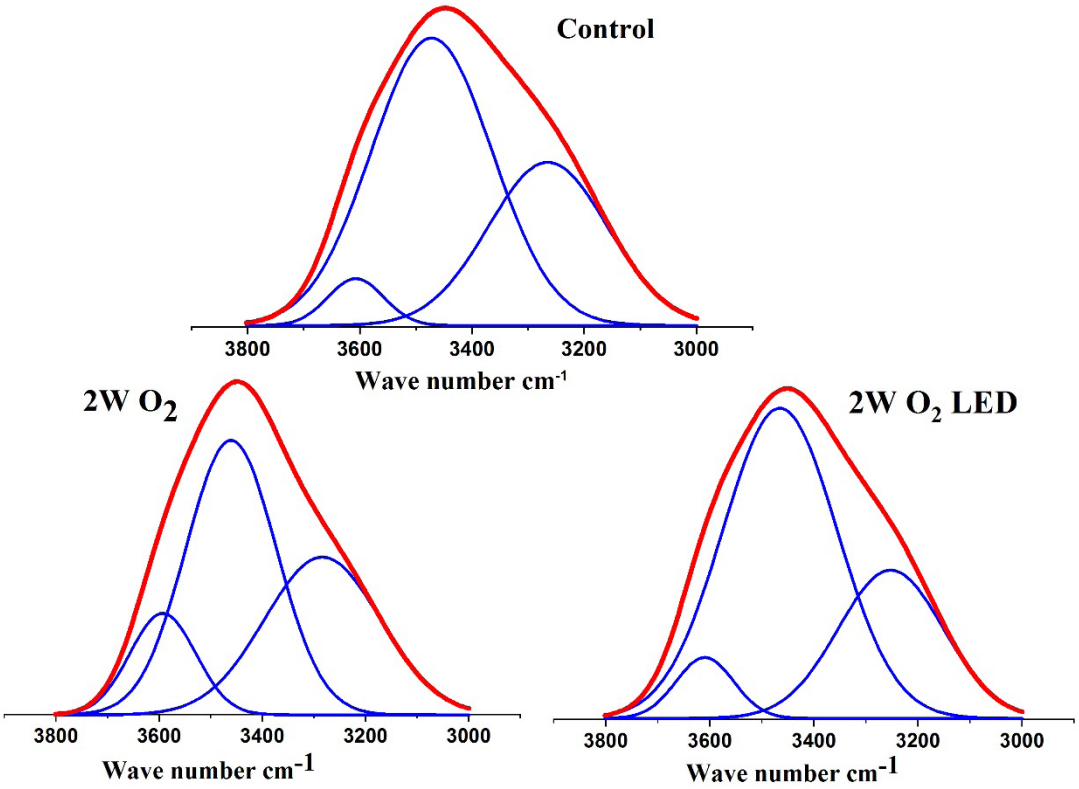

Fig.2. FTIR spectra of the control retina, after two weeks of hyperoxia for (2W O_2) and treated with LED laser (2W O_2 LED) groups in the NH-OH stretching region (3700- 3000 cm^{-1}).

CH stretching region

Figure (3), (4) showed the CH stretching region (3000-2800 cm^{-1}) for the retina of control, hyperoxia and LED laser treated groups after one and two weeks respectively. The profile of the control group revealed the presence of four peaks centered at 2961±1cm^{-1}, 2925±1cm^{-1}, 2877±1cm^{-1} and 3853±1cm^{-1} corresponding to CH_3 asymmetric, CH_2 asymmetric, and two peaks corresponding to frist and second CH_2 symmetric vibration modes, respectively (Coates 2000) . It was noticed that, both hyperoxia and LED laser treated groups, showed non significant changes ($p>0.05$) in the vibrational frequency and band widths after one and two weeks.

Fingerprint region

Figure (5) showed the fingerprint region (1000-1800 cm^{-1}) for control, after one week of hyperoxia and the hyperoxia group treated by low levle LED laser. The control group revealed the presence of 9 bands: (1) C=O stretching vibrations mode of carboxylic acid of the amino acid at 1727 cm^{-1}, (2) Amide I at 1650 cm^{-1}, (3) the amide II (NH bending and CH stretching) at 1545 cm^{-1}, (4) CH_2 bending at 1457 cm^{-1}, (5) COO_{sym} at 1401 cm^{-1} , (6) CH_3 deformation at 1294 cm^{-1} , (7) PO_2 asymmetric at 1230 cm^{-1}, (8) COO_{asym} at 1168cm^{-1}, (9) $_{sym}PO_2$ at 1083 cm^{-1}(Yang et al., 2007), (Aboualizadeh et al., 2017).

As shown in Table (3), in the group exposed to hyperoxia for one week, the vibrational frequency corresponding to ester C=O at 1727 cm^{-1} was shifted to1715 cm^{-1} ($p<0.05$) and its bandwidth increased. The amid I band at 1650 cm^{-1} was shifted to 1644 cm^{-1} ($p<0.05$) with no changes in its bandwidth. No significant change in the frequencies of the amide II band, COO_{sym} and $_{sym}PO_2$ but their bandwidths were increase. CH_3 deformtion band was significantly shifted from 1294 cm^{-1} to 1327 cm^{-1} ($p<0.01$), its bandwidth was significantly decreased. PO_2 asymmetric band vibrational mode that moved from 1229 cm^{-1} to 1234 cm^{-1} may have some increment in its bandwidth in addition to disappearance of COO_{asym} mode. After two weeks of hyperoxia , the same behavior of wave numbers and bandwidths were observed as in after one week of hyperoxia group.

After treatment with LED laser, the amide I frequency may be enhanced in both one week

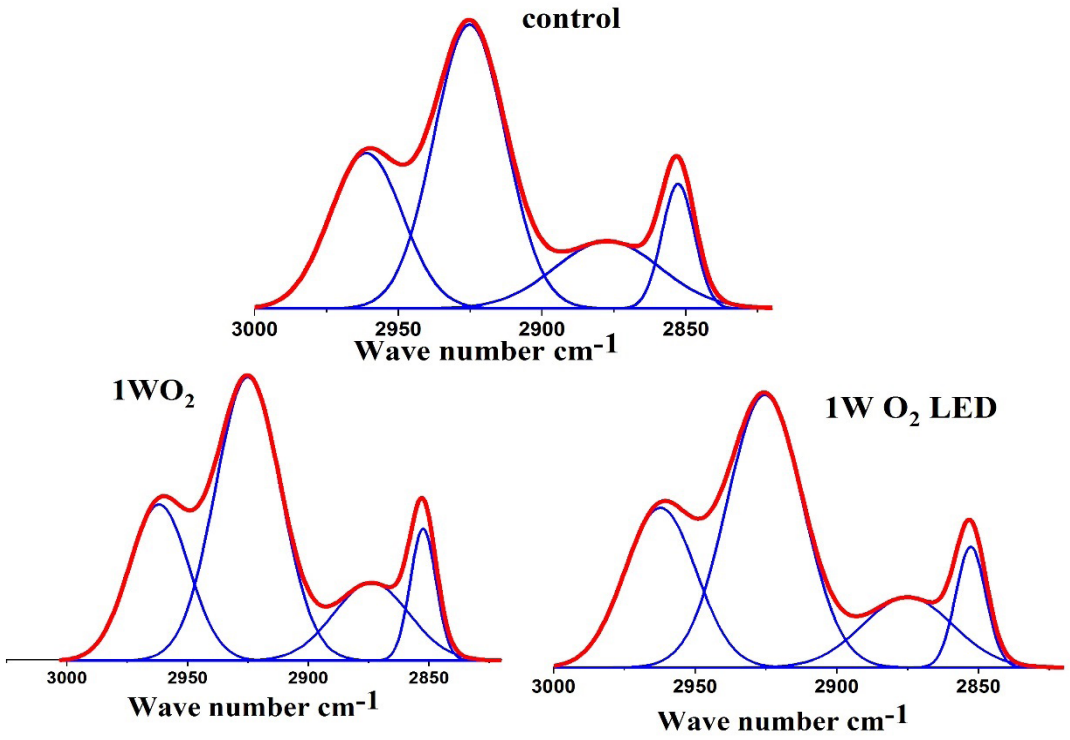

Fig. 3. FTIR for retina of control, hyperoxia for one week (1W O2) and LED laser treated (1W O2 LED) groups in CH stretching region (3000-2800 cm-1).

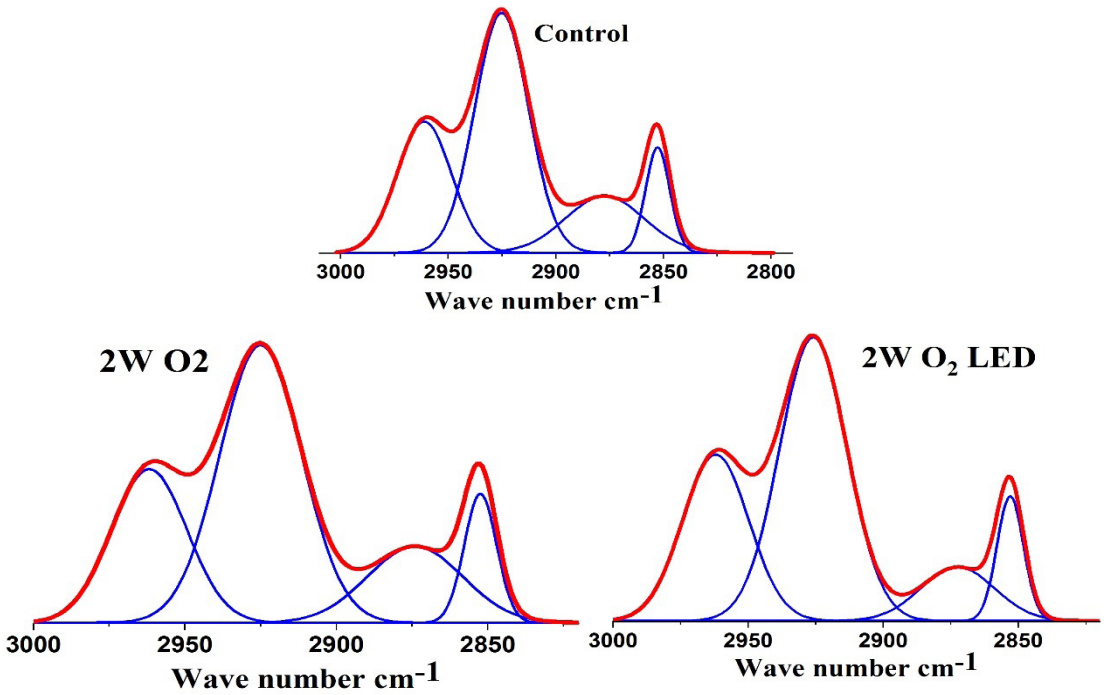

Fig. 4: FTIR for retina of control, hyperoxia for two weeks (2W O2) and LED laser treated (2W O2 LED) groups in CH stretching region (3000-2800 cm-1).

TABLE 2: Wavenumber and bandwidth of CH stretching region for all the studied groups after one and two weeks. The first line indicates wave number and the second for bandwidth.

Peak	Control	1W O_2	2W O_2	1W O_2 LED	2W O_2 LED
CH_3 asymmetric	2961±1 30±1	2961±1 29±1	2962±1 29±1	2962±1 30±1	2962±1 29±1
CH_2 asymmetric	2925±1 30±1	2925±1 32±1	2925±1 31±1	2925±1 31±1	2926±1 30±1
CH_2 symmetric	2877±1 44±2	2874±1 37±2	2874±1 32±2	2874±1 37±2	2872±1 39±3
CH_2 symmetric	3853±1 13±1	2852±1 13±1	2852±1 13±1	2853±1 13±1	2853±1 12±1

and two weeks with hyperoxia groups. The four bands of amide II, COO_{sym}, CH_3 Deform and PO_2 asymmetric have non-significant change in their vibrational frequencies ($p>0.05$), but they showed significant improvement in their bandwidths. The band at 1168 cm^{-1} assigned to COO_{asym} was appeared but with some changes in wavenumber and bandwidth.

Discussion

The purpose of this study is to report the efficacy of low level laser in treatment of retinal hyperoxia disorders in rats. The results in this study revealed that NH-OH region of FTIR spectra (Figure 1) of the retinal tissue is sensitive to systemic hyperoxia exposure, as shown in table (1). NH-OH region covers most of the membrane constituents contain NH bonds such as in proteins and lipids. Therefore, numerous distortions in the membrane structure of retinal cells may be produced due to exposure to 90% O_2. Moreover, the change in the vibrational frequencies of $_{str}O–H$ and $_{str}O–H_{sym}$ specified that the hydrogen bond has been destructed and/or weakened (Haris and Chapman 1994), (Yang et al., 2003) **.** This results agrees with previous studies that conducted on neonatal, adult rabbits and four months mice. This previous findings detected that exposure to high concentrations of O_2 is an important factors for emergence changes in the retinal protein structure, severe damage to photoreceptors with increasing oxidative stress, erosion of the blood-retina barrier (BRB) and loss of functional vision (Albarracin et al., 2013).

The CH stretching region of hyperoxial group can be considered with no changes, except slight narrowing in CH_{2sym} bandwidth. By analysis of the fingerprint region (Table 3), it was found that the exposure to high levels of O_2 encouraged obvious changes in the vibrational frequencies and bandwidths of some estimated components. Increasing in the bandwidth of C=O band and decreasing in its wavenumber, this may be related to the destruction of old H-bond and formation of new one. CH_3 Deform and PO_{2asym} bands have significant change in their bandwidths ($p<0.01$), this may lead to several variations in the nucleic acid content of phospholipids in hyperoxia group and spatial changes in the position of phosphate groups in protein helix . On the other hand, COO_{asym} band was missed after exposure to 90% O_2 which designated that molecular skeleton of protein has several fluctuations (Mahmoud et al., 2011).

After LED laser treatment, NHOH region results showed a clear improvement in all bands vibrational motion and bandwidths which may prove that LED laser exposure may lead to an improvement in the membrane structure of retinal cells. Another important feature in fingerprint region from rats treated with LED laser is that there are changes in the symmetrical and asymmetrical PO2 vibrations and bandwidths (Table 3) but they became better than those in hyperoxia group. Therefore, they probably evident initial stability in the secondary and tertiary structure of DNA [11]. Four bands of, amide II, CH2 bending,

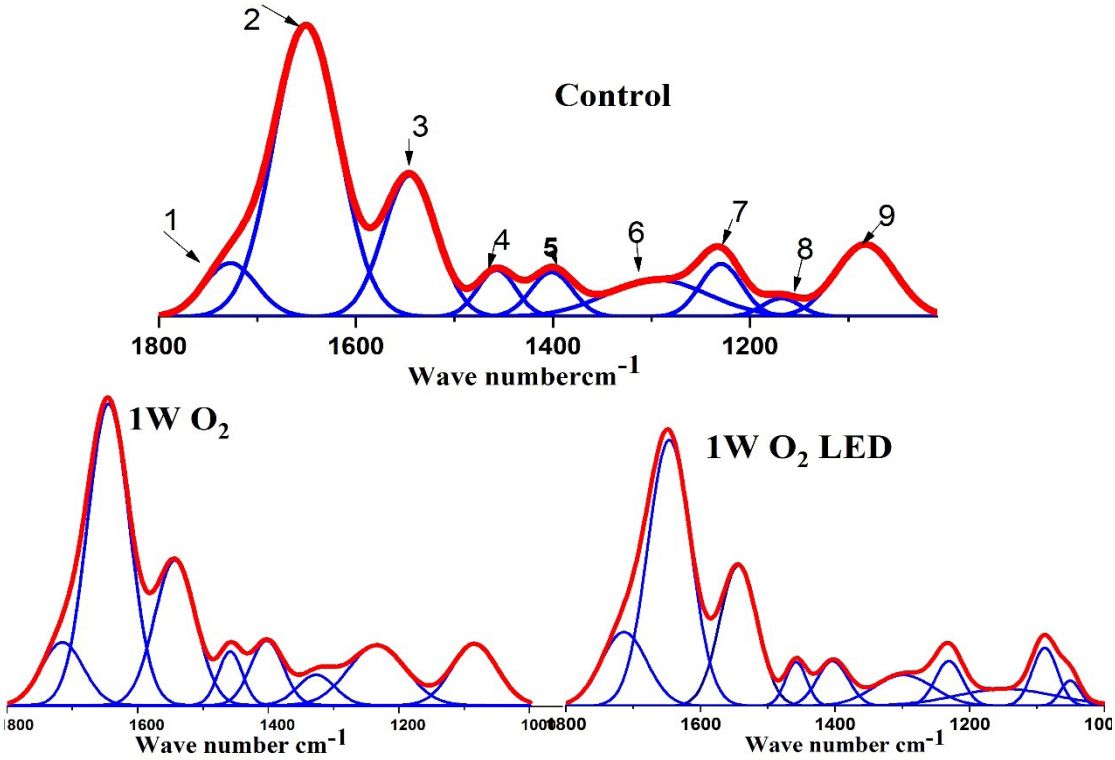

Fig.5. FTIR of rat's retina for normal, hyperoxia (1WO2) and LED laser treated groups (1W O2 LED) after 1 week in fingerprint region (1800–1000cm–1).

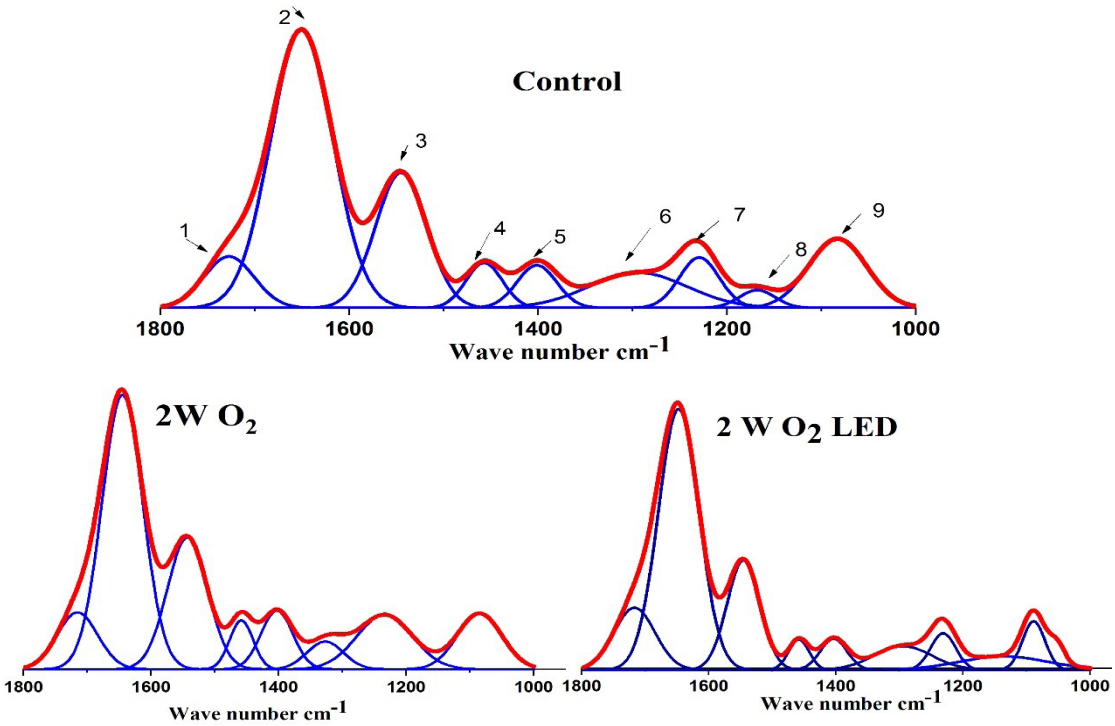

Fig.6. FTIR of rat's retina for normal, hyperoxia (2WO2) and LED laser treated groups (2W O2 LED) after 2 weeks in fingerprint region (1800–1000cm–1).

TABLE 3: Wavenumber and bandwidth of fingerprint region for control, (O_2) and group exposed to LED laser after 1 and 2 weeks

Peak	control	1W O_2	2W O_2	1WO_2 LED	2WO_2 LED
Ester C=O	1727±1 62±1	1715±2 76±1	1715±2 79±1	1714±1 75±1	1720±1 75±1
Amide I	1650±1 78±.5	1644±1 76±1	1645±1 76±1	1647±1 75±1	1647±1 76±1
Amide II	1545±1 64±1	1543±1 75±1	1543±1 75±1	1544±1 66±1	1545±1 61±1
CH2 bending	1457±1 46±1	1458±1 37±1	1458±1 37±1	1458±1 43±1	1458±1 40±1
CO_{Osy}m	1401±1 50±1	1403±1 59±1	1403±1 59±2	1404±1 54 ١±	1403±1 49 ١±
CH_3 Deform	1294±2 122±6	1327±1 67±2	1327±1 67±2	1298±1 104±4	1294±1 107±3
PO_{2asym}	1229±1 50±1	1234±1 103±1	1234±1 103±1	1231±1 47±1	1231±1 46±1
CO_{Oasy}m	1168±1 45±1	------	-------	1146±4 174±8	1136±3 164±6
$_{sym}P_O2$	1083±1 72±.5	1084±1 82±1	1084±1 82±	1088±1 45±1	1086±1 68±1

COO_{sym} and CH3 Deformation can be considered to have no changes in the vibrational frequency but small changes in bandwidths, in addition to reappearance of the band at 1086 cm^{-1} assigned for COO_{asym}. All these results may produce permanence in the confirmation of most of the fatty acids, phospholipids, triglycerides and cholesterol esters (Severcan et al., 2014).

LED laser treatment has a protective effects on the retina and this is may be related to mechanisms of action of low level laser therapy which is very different from the conventional use of photon energy in laser medicine where heating and burning are the usual mechanisms of action. Instead of depending on thermal activity, this new light therapy technique activates photochemical reactions of low-intensity FR/NIR (630-1000 nm) light with tissue. It was proposed that cytochrome C oxidase inside mitochondria serves as the primary photo acceptor for FR/NIR (630-1000 nm) light, the stimulation of cytochrome C oxidase by FR/NIR light is believed to lead to an increase in the energy production by mitochondria, protein synthesis, increase mitochondrial respiration and ATP synthesis (Geneva 2016).

These findings agreed with the findings of Albarcine et al (2013) who suggested that Photobiomodulation therapy is a treatment technique that has beneficial effects in most popular disease models for over 30 years including treatment of soft tissue injuries, diabetic wounds, radiation-induced problems, inflammation and neurodegenerative conditions. Moreover, 670 nm red light has sufficient ability to treat and/or reduce the progression of retinopathy and conditions involving cell death. This is due to its novel mechanism of action which can activate the production of transcription factor after light photon absorption, these factors responsible for protein synthesis, cell proliferation, anti-inflammatory signaling, anti-apoptotic proteins and antioxidant enzymes (De Freitas and Hamblin 2016).

Conclusion

According to our FTIR data, we could suppose that firstly, hyperoxia possibly induces modification in retinal protein structures and its genetic material through increasing the disordered and structural conformations in NH-OH and fingerprint regions. Furthermore, the present data

confirmed that irradiation with 670 nm LED laser ameliorates changes in membrane structure of retinal cells and protein structure. This is what has indicated by improvement in most of bands of the NHOH and fingerprint regions.

Refrences

1. Abdelkawi S. A., El Saeid A. A., El-Rashedi A. M., and Abd-Eldaiem M. S. Photobiomodulation therapy for diabetic macular edema: Fourier transform infrared spectroscopy study. Journal of The Arab Society for Medical Research, 13(1), 25 (2018).
2. Aboualizadeh E., Ranji M., Sorenson C., Sepehr R., Sheibani N., and Hirschmugl C Retinal oxidative stress at the onset of diabetes determined by synchrotron FTIR widefield imaging: towards diabetes pathogenesis Analyst, 142(7): 1061-1072.2017)).
3. Albarracin R., Natoli R., Rutar M., Valter K., and Provis J. 670 nm light mitigates oxygen-induced degeneration in C57BL/6J mouse retina. BMC neuroscience, 14(1), 125. (2013).
4. Coates J. Interpretation of infrared spectra, a practical approach. In: Encyclopedia of analytical chemistry. R. A. Mayer (Ed), Pp. 10815-10837, John Wileyand Sons Ltd, Chichester (2000).
5. De Freitas L. F., and Hamblin M. R. Proposed mechanisms of photobiomodulation or low-level light therapy. IEEE Journal of selected topics in quantum electronics, 22(3): 348-364. (2016).
6. Dovbeshko G. I, Gridina N. Y., Elena B, Kruglova E.B. and Pashchuk O.P. FTIR spectroscopy studies of nucleic acid damage. Talanta 53: 233-246. (2000).
7. Eells, J. T., Wong-Riley M. T., VerHoeve J., Henry M., Buchman E. V., Kane M. P. and Margolis D. Mitochondrial signal transduction in accelerated wound and retinal healing by near-infrared light therapy. Mitochondrion, 4(5), 559-567. (2004).
8. Geneva, I. I. Photobiomodulation for the treatment of retinal diseases: a review. International journal of ophthalmology, 9(1), 145. (2016).
9. Haris P. and Chapman D. Analysis of polypeptide and protein structures using Fourier transform infrared spectroscopy Methods Mol Biol; 22: 183–202. (1994).
10. Helmerhorst H. J., Arts D. L., Schultz M. J., van der Voort P.H., Abu-Hanna A., de Jonge E. and van Westerloo D. J. Metrics of arterial hyperoxia and associated outcomes in critical care. Crit Care Med;45(2):187–95(2017).
11. Mach W. J., Thimmesch A. R., Pierce J. T., and Pierce J. D. (2011). Consequences of hyperoxia and the toxicity of oxygen in the lung. Nursing research and practice, 2011.
12. Mahmoud S. S., El-Sakhawy E., Abdel-Fatah E. S., Kelany A. M., and Rizk R. M. Effects of acute low doses of Gamma-radiation on erythrocytes membrane. Radiation and environmental biophysics 50(1): 189-198. 2011)).
13. Rincon F., Kang J., Vibbert M., Urtecho J., Athar M. K., Jallo J. Significance of arterial hyperoxia and relationship with case fatality in traumatic brain injury: a multicentre cohort study. J Neurol Neurosurg Psychiatry;85(7): 799–805. (2014).
14. Severcan F., Bozkurt O., Gurbanov R. and Gorgulu G. FT-IR spectroscopy in diagnosis of diabetes in rat animal model. Journal of Biophotonics. 3(8-9): 621-631.2010)).
15. Yang L., Xu Y. and Su Y. Study on the variations of molecular structures of some biomolecules induced by free electron laser using FTIR spectroscopy. Nucl Instrum Methods Phys Res; 258:362–368. (2003).
16. Yang L., Xu Y., Su Y., Wu J., Zhao K., Wang M. and Chen J. E. Study on the variations of molecular structures of some biomolecules induced by free electron laser using FTIR spectroscopy. Nuclear Instruments and Methods in Physics Research Section B: Beam Interactions with Materials and Atoms, 258(2), 362-368.2007)).

العلاج بالليزر منخفض الطاقة لتشوهات بروتين الشبكية الناتجة عن فرط التأكسج والتي تم تقييمها بواسطة تحليل فوريير للأشعة تحت الحمراء

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قسم الفيزياء. كلية العلوم فرع البنات. جامعة الازهر- [2] قسم الطبيعه الحيوية معهد بحوث امراض العيون.[1] الجيزة

كان الهدف من هذه الدراسة هو النظر في الآثار الجانبية الخطرة للتعرض لتركيزات عالية من الأكسجين (فرط الأكسجه) على بروتين الشبكية وتقييم الدور الوقائى للعلاج بالليزر منخفض الطاقة. فى هذه الدراسة تم تقسيم ٥٠ فأر ابيض الى ثلاث مجموعات على النجو التالى (ا) المجموعة الظابطه (ن =١٠ فئران) دون التعرض لفرط الأكسجه ولا العلاج بالليزر (ب) مجموعة فرط الأكسجه (ن = ٢٠) تعرضت لتركيزات عاليه من الأكسجين ٩٠٪ يوميا لمدة أسبوع (ن =١٠) وأسبوعين (ن= ١٠) (ج) مجموعة فط التأكسج (ن =٢٠) حيث عولجت هذه المجموعة بالليزر الديودى منخفض الطاقة ٦٧٠ نانومتر بمعدل جلستين اسبوعيا . تم ذبح فئران التجارب لكل من مجموعات فرط التأكسج والعلاج بالليزر بعد أسبوع وأسبوعين وتم تطبيق تحليل فوريير لطيف الأشعه تحت الحمراء . أكدت نتائج تحليل بروتين الشبكية تأثرا واضحا للتعرض لفرط الأكسجه حيث ظهرت تشوهات

فقد أدت الى حيود الموجات عن مكانها وحدوث تغيرا(NH – OH and fingerprint)عديدة فى مناطق

ملحوظا فى سعة الموجات مما أظهر طفرات غريبة على الهيكل المعروف لبروتين الشبكيه . وباستخدام الليزر منخفض الطاقة ظهر تحسنا واضحا فى بروتين الشبكيه ويرجع ذلك لألية عمل هذا الضوء الذى يعمل على اثارة وتنشيط عوامل نسخ داخل الميتوكوندريا بعد امتصاص الفوتون . وهذه العوامل مسؤولة عن تخليق البروتين وتعويض التالف من الخلايا والتئام الجروح وارسال اشارات لمضادات الالتهابات والانزيمات المضادة للأكسده مما يعمل على تقليل اعتلال الشبكيه . وقد برهن تحليل فوريير على ذلك .