Iraqi JMS

Published by Al-Nahrain College of Medicine
P-ISSN 1681-6579
E-ISSN 2224-4719
Email: iraqijms@colmed-alnahrain.edu.iq
http://www.colmed-alnahrain.edu.iq
http://www.iraqijms.net
Iraqi JMS 2019; Vol. 17(2)

4-Hydroxy-2-nonenal, Induced Nitric Oxide Synthase Status in Hypertension with Kidney Disease Patients

Ali D. Hammed[1] *MSc*, **Alaa H. Jawad**[1] *PhD*, **Noor M. Ali**[2] *PhD*

[1]Dept. of Chemistry, College of Science, Al-Nahrain University, Baghdad, Iraq, [2]Dept. of Chemistry and Biochemistry, College of Medicine, Al-Nahrain University, Baghdad, Iraq,

Abstract

Background The oxidative stress is one of the main reasons for cardiovascular diseases and also one of results of these diseases, its development (like kidney disease).

Objective To identify the effect of oxidative stress nitric oxide and reactive oxygen species on cardiovascular diseases.

Methods The study involved 56 subjects comparable in age and sex divided into two groups; 28 hypertensives subjects with kidney disease and 28 apparently healthy subjects as control group. The following analysis was done: 4-Hydroxy-2-nonenal (4HNE), Induced nitric oxide synthase (iNOS) and albumin.

Results There was a significant increase in (4HNE) between patients' group and control group. iNOS was significantly higher in patients as compared to controls while there were no significant differences found in albumin between patient and control group. There is a positive relationship between oxidation results from hypertension and their developments. There is a positive correlation between BMI and disease.

Conclusion Based on this study is important on ideal weight, because obesity considered main factors for heart disease and hardening of the arteries. In addition, the effect of oxidative stress, which leads to high blood pressure and thus chronic kidney disease.

Keywords Chronic kidney disease, hypertension, 4-Hydroxy-2-nonenal (4HNE), Induced nitric oxide synthase (iNOS), albumin, creatinine, reactive oxygen species

Citation Hammed AD, Jawad AH, Ali NM. 4-Hydroxy-2-nonenal, induced nitric oxide synthase status in hypertension with kidney disease patients. Iraqi JMS. 2019; 17(2): 146-152. doi: 10.22578/IJMS.17.2.8

List of abbreviations: BMI, Body mass index, BP = Blood pressure, CKD = Chronic kidney disease, iNOS = Induced nitric oxide synthase, ROS =Reactive oxygen species

Introduction

Blood pressure (BP) is the force created by the heart as it pushes blood into the arteries through the circulatory system. Every time the heart contracts or beats, the blood is pumped out and creates a wave of pressure in the arteries. Macrophages had been supposed to be the source of the most reactive oxygen species (ROS) in the vessel's wall [(1)]. However, it has become clear that all the cells in the vessel wall produce ROS in different quantities and in response to diverse stimuli [(2)]. High BP is one of the main factors causing the disease, which contributes to the deterioration of kidney function [(2)]. The presence of kidney disease is a medical reason by the subscriber to find and underappreciated from high BP resistance. Therefore, treatment of hypertension has become the most important intervention in the management of all type of chronic kidney disease (CKD) [(3)]. CKD

CC BY NC ND

(also called chronic kidney failure) is a condition that affects the function of the kidney, which can progress over time to kidney failure. When the kidneys fail, dialysis or a kidney planting is needed to support life. Many diseases can cause CKD [4]. The most common are diabetes and high BP. High BP puts more pressure on the blood vessels throughout the body, including the kidney filters (nephrons). Hypertension is the number two cause of kidney failure [5]. In addition, increased oxidative stress causes tissue damage by different mechanisms including promoting lipid peroxidation, DNA damage, and protein modification [6]. ROS is a group which are either free radicals or molecular species capable of generating free radicals, examples include singlet oxygen, superoxide, peroxides, and hydroxyl radical. In a biological context, ROS which is formed as a naturally byproduct of the natural metabolism of oxygen and have important roles in cellular signal and homeostasis, but through times of environmental stress (like, UV or heat exposure) ROS levels can raise dramatically [7], and this may lead to severe damage to cell structures, Cumulatively, this is called oxidative stress. ROS are also created by exogenous sources like ionizing radiation [8]. When there is an imbalance between the generation of ROS and the antioxidant defense system so that the last one becomes overwhelmed, oxidative stress happens [2]. Oxidants and free radicals are produced inevitably through most physiological and metabolic processes, and the human body has an anti-oxidant defense mechanisms, these different mechanisms according to the type of cells and tissues, and they may behave aggressively or synergy [8]. Oxidative stress causes many pathophysiological conditions in the body [9], including neurodegenerative diseases such as Alzheimer's disease, gene mutations, chronic fatigue syndrome, atherosclerosis and inflammatory diseases [10]. In physiological conditions, low level ROS play a role in the protection the organism, while high levels of ROS may cause damage to the structures of the cell, nucleic acids, lipids, proteins or DNA damage [11]. 4-hydroxy-2-nonenal (4HNE), a high toxicity product of lipid peroxidation, is an inhibitor of mitochondrial respiration. 4HNE exerts its influence on respiration by inhibiting α-ketoglutaratedehydrogenase (KGDH) [12]. A study by Campos et al. in 2015 observed an increase in 4HNE with hypertension and kidney disease [13]. Nitric oxide synthases (NOSs) are a family of enzymes catalyzing the production of nitric oxide (NO) from L-arginine [14].

Albumin is the most abundant protein in the circulatory system and act as antioxidant [15], the antioxidant activity of albumin due to its ability to bind bilirubin, homocysteine and lipids [16].

The objective of the study was to investigate the relationship between of some oxidative stress markers and kidney disease associated with hypertension.

Methods

The present study comprised of 56 subjects divided into two groups control group (28) and hypertensive with kidney disease group (28) aged between 22-65 years. These patients were hospitalized at Educational Laboratories in the Al-Yarmouk Teaching Hospital. Blood sample were collected and centrifuged at [4000 xg] for 5 min after clotting. The resultant serum were separated and stored at [-20] °C until used. Estimation of serum albumin was done using kit provided by Bio Systems Company.

Also, Creatinine in the blood has been estimated using several provided by Randox. Serum 4HNE is typically quantified from serum samples with the most popular method being a colorimetric assay based on biotin double antibody sandwich technology. The serum INOS is typically quantified from the serum test samples employed by the quantitative quantification enzyme immunoassay technique.

CC BY NC ND

Statistics

The Statistical Analysis System-SAS- (2012) was used to determine of various factors in studied parameters.

Results

Fifty-six sample patients comprising of 28 patients and 28 apparently healthy were included in the present study, table 1 shows the means and standard deviation of age, body mass index (BMI), duration of disease, 4HNE, in addition to albumin levels and creatinine levels for the control and patients' group.

Table 1. Characteristics of the Hypertension (HT) and control group (mean ± SD)

Characteristic	Hypertension with kidney group n=28	Control group n=28	P-Value
Age (year)	47.60 ± 11.71	23.82 ± 5.02	<0.01
Body Mass index (Kg/m^2)	28.17 ± 3.90	25.18 ± 2.89	<0.01
Systolic blood pressure (mmHg)	19.03 ± 3.23	11.96 ± 0.66	<0.01
Diastolic blood pressure (mmHg)	10.83 ± 1.25	8.63 ± 0.56	<0.01
4HNE (ng/L)	273.71 ± 96.75	136.46 ± 24.55	<0.01
iNOS (IU/ml)	15.58 ± 14.44	6.42 ± 4.93	<0.01
Albumin (g/dl)	3.90 ± 0.48	4.73 ±0.25	<0.01
Creatinine (mg/dl)	8.01 ± 5.88	0.75 ± 0.09	<0.01

The highly significant difference ($p<0.01$) of 4HNE in HT with CKD patient as shown in figure 1 may be due to high oxidative stress which generally causes damage to the membrane polyunsaturated fatty acids which leading to the generation of 4HNE as shown in figure 2 and 3 and this result agrees with study done by Usberti et al. in 2002 [21].

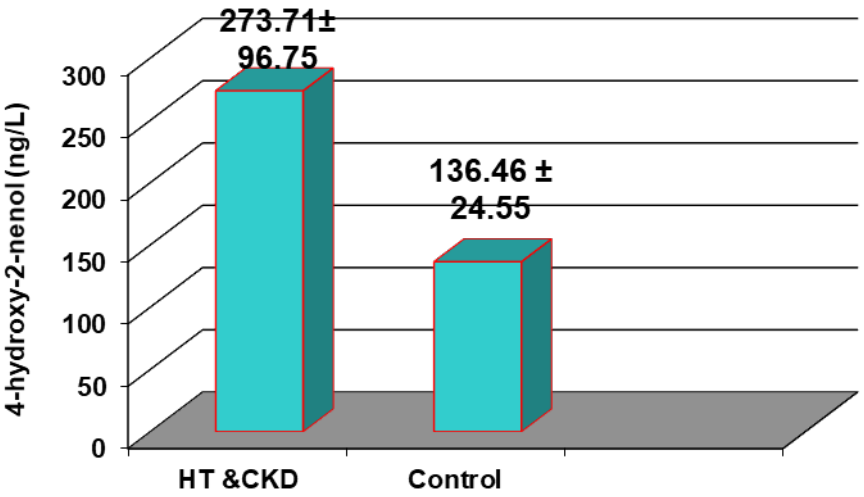

Figure 1. Mean concentration (ng/L) of oxidative marker 4HNE for studied groups

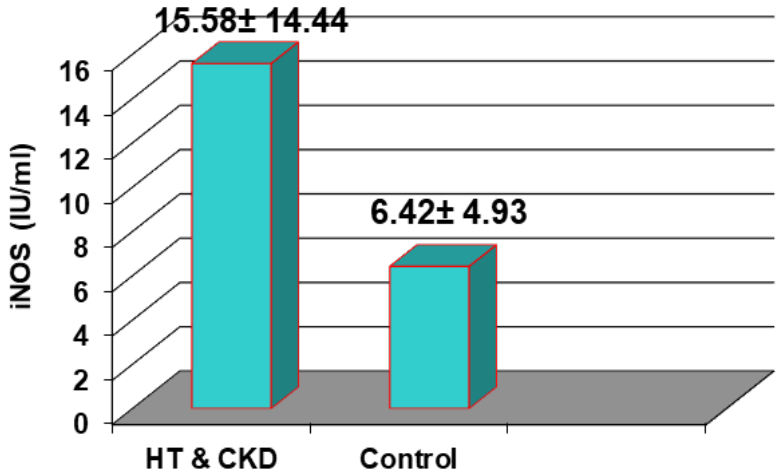

Figure 2. Mean concentration (IU/ml) of INOS in the studied groups

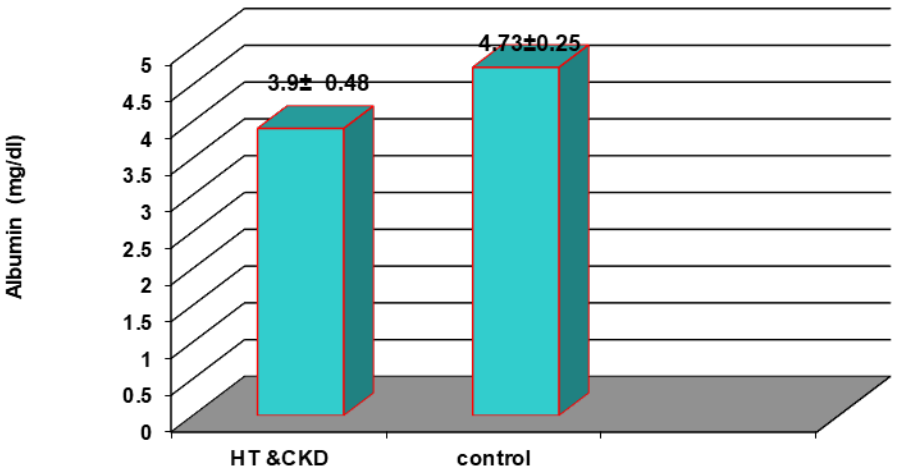

Figure 3. Mean concentration (g/dl) of albumin in the studied groups

Discussion

There is a significant difference ($p<0.01$) in age when comparing patients' groups with control group. The significant difference in BMI ($p<0.01$) between HT patients with CKD patients and control group reveal the positive correlation between obesity and disease. This finding was similar to Hall. et al in 2014 who found a significant increase in BMI in hypertensive with kidney disease patients [19]. Obese patients are more able to be hypertensive than lean patients, and weight gain is usually associated with increases in arterial pressure [20].

The results of this study showed a significant difference in iNOS between hypertension with kidney patient and control groups ($p<0.01$). In this study, results were agreed with Abd El Gawad et al. in 2011 [22] who found a significant difference in iNOS levels in hypertensive patients. There is a positive correlation between serum iNOS levels and systolic, diastolic and mean blood pressure. Several studies have demonstrated high iNOS level in patients with hypertension [23].

Impaired endothelial dependent vasodilatation associated with abnormal iNOS may be an important factor in the development and progression of atherosclerosis and hypertension. In addition, endothelial dysfunction, in hypertensive patients may initiate vascular inflammation that leads to cytokine-induced activation of inducible NOS which favors the formation of peroxynitrite contributing to cytotoxicity and tissue injury [11].

Excessive or inappropriate NO production by iNOS reacts with superoxide anions in the plaque to yield reactive oxidant species such as peroxynitrite contributing to cytotoxicity and tissue injury. Another possible outcome for NO coming from iNOS could be the activation of matrix metalloproteinase and induction of apoptosis in smooth muscle cells and macrophages.

Albumin contains a free sulfhydryl group, and this forms a disulfide with many compounds like cysteine, homocysteine, or glutathione, Albumin is able to scavenge hydroxyl radicals [23], the decrease in albumin in patient is agreed with results of Menon et al. in 2005 in hypertension [17] who suggested that result is may be due to its function as antioxidant activity. On the other hand, the non-oxidized albumin is decrease in addition to negative acute phase protein, so inflammation is considered the main cause of a decline in the serum albumin [23]. The results showed a significant in creatinine levels in patients' groups when compared to control group and this agree with AL-Hamdani [24]. There were significant differences between patients and

control in creatinine concentration This elevation of serum creatinine concentration may be due to the decline in creatinine clearance due to the lack in the GFR [25]. Creatinine as a waste product of creatine produced in muscles, and is converted about 1% of the total pool muscle creatine and creatinine daily through the loss of water is enzymatically spontaneous. Since released in the blood at a constant rate, and since it matched closely to the secretion of glomerular filtration rate [26]. The elevation of serum creatinine concentration may be attributed to the lack in creatinine clearance due to the decline in the GFR.

The weak negative correlation between 4HNE and albumin in hypertension with kidney patient and control group agreed with Aldini et al. in 2006 who found a negative correlation between 4HNE and albumin [27]. There is an evidence for a significant antioxidant activity of the represent the major and predominant circulating antioxidant in plasma known to be exposed to continuous oxidative stress.

There is also negative correlation between iNOS and albumin in patient and control group this result shown Poteser and Wakabayashi in 2004 who found a negative correlation between iNOS and Albumin [28]. There is a negative correlation between 4HNE and creatinine in patient and control group was observed in this study which agreed with Kobori et al. who found a negative correlation between 4HNE and creatinine [29]. A negative correlation between iNOS and creatinine in patient and control group was observed in this study which agreed with Park et al. who found a negative correlation between iNOS and creatinine [30].

Based on this study is important on ideal weight, because obesity considered main factors for heart disease and hardening of the arteries. In addition, the effect of oxidative stress, which leads to high blood pressure and thus chronic kidney disease.

Acknowledgement

The authors would like to thank Al-Nahrain University and Al-Yarmouk Teaching Hospital for the financial supports and research facilities.

Author contribution

Hammed: conducted the sampling, work and writing. Dr. Jawas and Dr. Ali supervised the work.

Conflict of interest

The authors declare no conflict interest

Funding

Self-funding.

References

1. Yancy CW, Jessup M, Bozkurt B, et al. 2016 ACC/AHA/HFSA Focused update on New pharmacological therapy for Heart Failure: An update of the 2013 ACCF/AHA Guideline for the Management of Heart Failure. A Report of the American College of Cardiology/American Heart Association task Force on Clinical practice Guidelines and the Heart Failure Society of America. Circulation. 2016; 134: e282-e293. doi: 10.1161/CIR.0000000000000435.
2. Ahmad A, Singhal U, Hossain MM, et al. The role of the endogenous antioxidant enzymes and malondialdehyde in essential hypertension. J Clin Diagn Res. 2013; 7(6): 987-90. doi: 10.7860/JCDR/2013/5829.3091.
3. de Jong MR, Hoogerwaard AF, Gal P, et al. Persistent increase in blood pressure after renal nerve stimulation in accessory renal arteries after sympathetic renal denervation. Hypertension. 2016; 67(6): 1211-7. doi: 10.1161/HYPERTENSIONAHA.115.06604.
4. Martínez-Castelao A, Górriz JL, Segura-de la Morena J, et al. Consensus document for the detection and management of chronic kidney disease. Nefrologia. 2014; 34(2): 243-62. doi: 10.3265/Nefrologia.pre2014.Feb.12455.
5. Köttgen A, Glazer NL, Dehghan A, et al. Multiple loci association with indices of cardivascular disease J. Acta Nat Genet. 2009; 41(6): 712-7. doi: 10.1038/ng.377.
6. Bhale DV, Hivre MD, Mahat RH, et al. Study of oxidative stress in patients with hypertension. Int J Recent Trends Sci Technol. 2013; (9): 157-8.
7. Palmer RM, Ferrige AG, Moncada S. Nitric oxide release accounts for the biological activity of endothelium-derived relaxing factor. Nature. 1987; 327(6122): 524-6. doi: 10.1038/327524a0.
8. Morales-González JA. Oxidative Stress and chronic degenerative disease a role for antioxidants. Rijeka, Croatia: Janeza Trdine; 2013; doi: http://dx.doi.org/10.5772/45722.

9. Ramalingam M, Kim SJ. Reactive oxygen/nitrogen species and their functional correlations in neurodegenerative diseases. J Neural Transm (Vienna). 2012; 119(8): 891-910. doi: 10.1007/s00702-011-0758-7.
10. Chandra K, Salman SA, Mohd A, et al. Protection against FCA induced oxidative stress induced DNA damage as a model of arthritis and in vitro anti-arthritic potential of Costus speciosus Rhizome Extract. Int J Pharmacog Phytochem Res. 2015; 7(2): 383-9. doi: 10.1186/s12906-016-1403-7.
11. Fliser D, Kronenberg F, Kielstein JT, et al. Asymmetric dimethylarginine and progression of chronic kidney disease: the mild to moderate kidney disease study. J Am Soc Nephrol. 2005; 16(8): 2456-61. doi: 10.1681/ASN.2005020179.
12. Humphries KM, Szweda LI. Selective inactivation of α-ketoglutarate dehydrogenase and pyruvate dehydrogenase: reaction of lipoic acid with 4-Hydroxy-2-nonenal. Biochemistry. 1998 Nov 10;37(45):15835-41. doi: 10.1021/bi981512h.
13. Campos JC, Fernandes T, Bechara LRG, et al., Increased clearance of reactive aldehydes and damaged proteins in hypertension-induced compensated cardiac hypertrophy: impact of exercise training. Oxid Med Cell Longevity. 2015; 2015, Article ID 464195, 11 pages, doi: https://doi.org/10.1155/2015/464195.
14. Elremaly W, Mohamed I, Rouleau T, et al. Adding glutathione to parenteral nutrition prevents alveolar loss in newborn guinea pig. Free Radic Biol Med. 2015; 87: 274-81. doi: 10.1016/j.freeradbiomed.2015.06.040.
15. Taverna M1, Marie AL, Mira JP, et al. Specific antioxidant properties of human serum albumin, a Review. Ann Intensive Care. 2013; 3(1):4. doi: 10.1186/2110-5820-3-4.
16. Oda E. Decreased serum albumin predicts hypertension in a Japanese health screening population. Intern Med. 2014; 53(7): 655-60. doi: 10.2169/internalmedicine.53.1894.
17. Menon V, Greene T, Wang X, et al. C-reactive protein and albumin as predictors of all-cause and cardiovascular mortality in chronic kidney disease. Kidney Int. 2005; 68(2): 766-72. doi: 10.1111/j.1523-1755.2005.00455.x.
18. AL-Hamdani IH. Estimation of Serum uric acid, urea and creatinine in essential hypertensive patients. Tikrit Med J. 2010; 16(1): 152-8.
19. Hall ME, do Carmo JM, da Silva AA, et al. Obesity, hypertension, and chronic kidney disease. Int J Nephrol Renovasc Dis. 2014; 7: 75-88. doi: 10.2147/IJNRD.S39739.
20. Lavie CJ, Milani RV, Ventura HO. Obesity and cardiovascular disease risk factor, paradox, and impact of weight loss. J Am Coll Cardiol. 2009; 53(21): 1925-32. doi: 10.1016/j.jacc.2008.12.068.
21. Usberti M, Gerardi GM, Gazzotti RM, et al. Oxidative stress and cardiovascular disease in dialyzed patients, Nephron. 2012; 91(1): 25-33. doi: 10.1159/000057601.
22. Abd El Gawad SS, Yossof MM, Soliman AAW, et al. Expression of inducible nitric oxide synthase gene in diabetic and non-diabetic coronary artery disease patients. Researcher, 2011; 3(6): 40-8.
23. Roche M, Rondeau P, Singh NR, et al. The antioxidant properties of serum albumin. FEBS Letters. 2008; 582: 1783-7. doi: https://doi.org/10.1016/j.febslet.2008.04.057.
24. Koyner JL, Vaidya VS, Bennett MR, et al. Urinary biomarkers in the clinical prognosis and early detection of acute kidney injury. Clin J Am Soc Nephrol. 2010; 5(12): 2154-65. doi: 10.2215/CJN.00740110.
25. Fox SI. Human Physiology. 8th ed. New York, NY: McGraw Hill; 2004. p. 406-39.
26. Patel P, DiPette DJ. Hypertension-related congestive heart failure in west Africa: a framework for global blood pressure control. J Clin Hypertens (Greenwich). 2015; 17(4): 260-2. doi: 10.1111/jch.12494.
27. Aldini G, Gamberoni L, Orioli M, et al. Mass spectrometric characterization of covalent modification of human serum albumin by 4-hydroxy-trans-2-nonenal. J Mass Spectrom. 2006; 41(9): 1149-61. doi: 10.1002/jms.1067.
28. Poteser M, Wakabayashi I. Serum albumin induces iNOS expression and NO production in RAW 267.4 macrophages. Br J Pharmacol. 2004; 143(1): 143-51. doi: 10.1038/sj.bjp.0705897.
29. Kobori H, Katsurada A, Ozawa Y, et al. Enhanced intrarenal oxidative stress and angiotensinogen in IgA nephropathy patients. Biochem Biophys Res Commun. 2007; 358(1): 156-63. doi: 10.1016/j.bbrc.2007.04.105.
30. Park KM, Byun JY, Kramers C, et al. Inducible nitric-oxide synthase is an important contributor to prolonged protective effects of ischemic preconditioning in the mouse kidney. J Biol Chem. 2003; 278(29): 27256-66. doi: 10.1074/jbc.M301778200.

Correspondence to Dr. Noor M. Ali
E-mail: noormustafali@yahoo.com
dr.noor.ali@colmed-alnahrain.edu.iq
Received Jul. 8th 2017
Accepted Jun. 27th 2019