Mini Review
Open Access
CrossMark

Investigating inexpensive treatments altering the nitric oxide system for obesity-related type 2 diabetes and diabetic nephropathy

Volume 2 Issue 2 - 2016

Sharon Inman R, Sonja Porter M
Department of Biomedical Sciences, Ohio University, USA

Correspondence: Sharon Inman R, Department of Biomedical Sciences, Ohio University, Athens, OH 45701, USA, Tel 017405932936, Email inmans@ohio.edu

Received: December 17, 2015 | **Published:** February 12, 2016

Abbreviations: T2DM, type 2 diabetes mellitus; NO, nitric oxide; NBC, niacin-bound chromium; LA, l-arginine; NOS, nitric oxide synthase; cGMP, cyclic guanosine monophosphate; GFR, glomerular filtration rate

Introduction

Diabetic nephropathy has become a leading cause of end stage renal disease with nearly 500,000 U.S. cases in the last ten years. Current, traditional pharmacologic approaches are costly and largely ineffective. While diet is crucial to the development of obesity and equally significant in the control of diabetes, the potential application of dietary supplements as a therapeutic intervention targeted to components of the diabetic nephropathy disease process has remained virtually unexplored. An efficacious, inexpensive, carefully targeted dietary approach to decrease or delay diabetic nephropathy in type 2 diabetes mellitus (T2DM) patients is highly possible and merited. Development of targeted intervention tools requires an appreciation of the underlying mechanism (s) of the pathology related to diabetic nephropathy. Because the pathophysiology is complex and involves many factors, the mechanisms involved in the development of diabetic nephropathy have not been fully characterized. However, increased cellular oxidative stress and alterations in the nitric oxide (NO) pathway have been shown to be involved in diabetic nephropathy. Hence, our laboratory has studied the effectiveness of effectiveness of L-arginine supplementation, the effectiveness of antioxidant (AO) diets and the role of oxidative stress, and administration a niacin-bound chromium diet (NBC) on obesity-related Type 2 diabetic rats to determine the effects on renal function and on alterations of the NO pathway.[1,2]

L-arginine supplementation

The nitric oxide (NO) system has been shown to be altered in diabetes and in diabetic nephropathy.[3] Nitric oxide is a vasodilator and if it is deficient or its metabolism is altered, this affects renal function and insulin sensitivity.[4] The precursor for NO is L-arginine (LA). LA is an amino acid that is synthesized within the body and can also be found in various types of food. LA is converted to NO by nitric oxide synthase (NOS).[5] Endothelial NOS (eNOS) results in NO release from the endothelium of blood vessels and causes vasodilation via cyclic guanosine monophosphate (cGMP).[6] Inducible NOS (iNOS) is an isozyme that is present in an oxidative environment. High levels of iNOS produce larger amounts of NO, which allows NO to react with superoxide forming peroxynitrite and thus leads to cell toxicity and/or death.[7] Therefore, higher levels of renal eNOS compared to iNOS would be beneficial in the late stages of diabetic nephropathy to maintain renal blood flow via vasodilation. While it is true in the early stages of diabetic nephropathy that there is renal vasodilation and hyper filtration, it is in the later stages that glomerular filtration rate decreases and the continued availability of nitric oxide would maintain glomerular filtration rate and renal blood flow.[8] Our hypothesis was that nitric oxide bioavailability may be increased by supplemented L-arginine dietary treatment in the later stages of diabetic nephropathy. It is upregulated by endothelial nitric oxide synthase rather than by the arginase enzyme. LA deficiency causes endothelial inflammation and cardiovascular disorders, and dietary LA supplementation can reverse these disorders.[9,10] In patients with type 2 diabetes mellitus (T2DM), LA supplementation resulted in a significant increase in NO concentration and total antioxidant status of the patient.[11] In a rat model of T2DM, the obese Zucker rat, it has been reported these rats had significantly lower renal function compared to the diabetic animals fed an antioxidant diet. This suggests that in an oxidative stress environment renal function declines and may be due to the increased production of peroxynitrite via iNOS resulting in cell death.[2]

Our studies utilized the obese Zucker rat (fa/fa rat) which exhibits hyperinsulinemia and hyperlipidemia as early as four to six weeks of age.[3,12] By six weeks of age the oral glucose tolerance test is abnormal in the obese fa/fa rat compared with its lean littermates, Fa/Fa.[4] Obese (fa/fa) Zucker rat is a spontaneous genetic obesity model which lacks the leptin receptor and exhibits hyperplasia, hyperinsulinemia, and hyperlipidemia. Its lean littermate Zucker rat (Fa/Fa) does have the leptin receptor and is used as the non-diabetic control for the obese Type 2 Zucker rat. LA supplementation resulted in higher GFRs compared to the Nondiabetic Control rats and at weeks 3 and 8 compared to Diabetic Controls. GFRs in the Nondiabetic Control rats were lower than those in the treated Diabetic Control rats at weeks 0 and 3 but were higher at weeks 6 and 8. Western blots were performed to assess the renal cortex and medulla protein levels of eNOS and iNOS. The eNOS protein levels were higher in both the renal cortex and medulla in LA-treated diabetic rats compared to untreated diabetic rats. Likewise, the iNOS protein levels were lower in both the renal cortex and medulla in LA-treated diabetic rats compared to untreated diabetic rats. Increased levels of eNOS and lower levels of iNOS in the renal medulla and cortex were detected in the LA supplemented rats compared to the diabetic control rats. eNOS causes positive activation of NO leading to cGMP and vasodilation.[13] iNOS causes toxification of NO forming peroxynitrite which leads

to cell damage and death.[14] Increased urinary cGMP levels in LA supplemented rats were observed. This is most probably due to an overabundance of plasma cGMP being produced from eNOS in the NO pathway. L-arginine may be an inexpensive alternative treatment for type 2 diabetics. In our study, early intervention with L-arginine supplementation was beneficial by preserving glomerular filtration rates, presumably via increased renal endothelial nitric oxide synthase levels leading to renal vasodilation; however, additional studies are needed to examine the alterations in the other many mediators involved in the nitric oxide pathway to further support this claim. Lastly, there was an improvement in the insulin sensitivity in the LA treated diabetic rats versus untreated diabetic rats.

Antioxidant diet administration

Diabetic nephropathy is also associated with oxidative stress and changes in renal nitric oxide (NO) production[15] which leads to endothelial dysfunction and decreased renal perfusion. Circulating levels of nitrite and nitrate (NOx) are increased in patients with T2DM and obese subjects compared with lean, healthy individuals.[16] In addition, alterations in urinary nitric oxide (NO) production and utilization can result in NO interaction with O2− to become peroxynitrite which causes Cytotoxicity.[17] Hypertension associated with oxidative stress may further aggravate the glomerular dysfunction.[18] Results of treatment of human diabetes with antioxidants in the diet have been encouraging but variable.[17] However, oxidative imbalance may start well before there is evidence of overt nephropathy and some studies have shown a benefit of antioxidants on glycemic control and the development of diabetic vascular complications.[17,19] When antioxidants are started on the day of streptozotocin induction of T1DM in rats, renal function is preserved,[20] and early treatment with vitamins C and E plus insulin has been shown to significantly lower blood glucose in this rat model.[21] Therefore it is important to attempt to develop a dietary antioxidant therapy that will work in T2DM as well. Supplementation with antioxidants and factors essential to nitric oxide (NO) production may have the potential to improve endothelial dysfunction and renal perfusion in T2DM.[22] Our strategy was to begin antioxidant therapy at an age prior to the appearance of renal dysfunction (4weeks), to determine if early intervention also works in type 2diabetes. We evaluated renal structure and function as well as associated metabolic factors including blood glucose and insulin, mean arterial pressure, urinary albumin and NO levels. As the animal's age and the diabetes progresses, both insulin resistance and blood pressure become more severe. Also as insulin resistance worsens and renal damage becomes more apparent, urinary albumin increases. in obese Zucker rats administered the antioxidant supplemented diet (13 and 20weeks of age) had higher glomerular filtration (GFR) rates, improved insulin resistance and higher urinary NO levels compared to untreated obese Zucker rats.

Niacin-bound chromium supplementation

Research has been done based on how perturbations in glucose/insulin metabolism are associated with enhanced lipid peroxidation secondary to greater free radical formation. Free radicals of oxygen are important known causes of tissue damage and have been associated with many aspects of aging including inflammatory diseases, cataracts, diabetes, and cardiovascular diseases. Augmented free radical formation and lipid peroxidation are not uncommon in diabetes mellitus, commonly associated with "premature aging". Ingestion of sugars, fats, and sodium have been linked to decreased insulin sensitivity, while caloric restriction, exercise, ingestion of chromium, vanadium, soluble fibers, magnesium, and certain antioxidants are associated with greater insulin sensitivity. Thus, manipulation of diet by influencing the glucose/insulin system may favorably affect lifespan and reduce the incidence of chronic disorders associated with aging.[23] We tested the effectiveness of NBC in slowing cellular damage due to alterations in the nitric oxide pathway in rats that have diabetic nephropathy due to type II diabetes mellitus (T2DM). Rat studies demonstrated that T2DM decreases both the production and bioavailability of nitric oxide (NO), and we hypothesized that NBC supplementation will preserve NO production via increased endothelial nitric oxide synthase (eNOS) and decreased neuronal nitric oxide synthase (nNOS). NBC supplementation displayed decreased levels of plasma creatinine in the treated diabetic rats compared to the untreated diabetic rats, and therefore preserved glomerular filtration rate (GFR). We provide evidence that this effect may also be due to increased levels of eNOS, which leads to renal microvascular vasodilation. Supplementation of NBC also showed lower nNOS levels, which is a significant finding when considering the role of nNOS in T2DM. Marked improvements in fasting glucose and lowered body weights were observed in the treated diabetic rats compared to the untreated diabetic rats. Therefore, these results demonstrate that NBC slows the progression of diabetic nephropathy in T2DM rats. However more research should be performed specifically focusing on alterations in the NO pathway with this supplemented diet in obesity-related type 2 diabetes and diabetic nephropathy.

Conclusion

Intensive insulin therapy may delay DN by five years if diabetes onset is at age 15, however renal failure still presents in 33% of patients by age 45.[24,25] Insulin therapy is costly and there is insufficient evidence to show that intensive glycemic control using insulin therapy reduces the risk for significant clinical renal outcomes.[26] This research is relevant since renal disease (nephropathy) resulting from obesity and type 2 diabetes is the leading cause of chronic renal failure and is a significant financial burden for maintaining patients on dialysis, and support through transplantation. The goal of these studies was to determine if L-arginine, an antioxidant diet and niacin-bound chromium supplementation can alleviate and/or abolish nitrosative and oxidative stress in the kidney that contributes to diabetic nephropathy. These dietary supplements could provide a relatively inexpensive treatment to help slow or prevent the progression of chronic diabetic kidney disease. This will vastly improve the quality of life for individuals with diabetic kidney disease. Thus, these studies to decrease nitrosatve stress, oxidative stress and preventing alterations in the NO pathway should lead to more studies and hopefully in the future, reduce the medical and socioeconomic burdens of human disability.

Acknowledgements

None.

Conflict of interest

Author declares that there is no conflict of interest.

References

1. Claybaugh T, Decker S, McCall K, et al. L-Arginine Supplementation in Type II Diabetic Rats Preserves Renal Function and Improves Insulin Sensitivity by Altering the Nitric Oxide Pathway. *Int J Endocrinol.* 2014;2014:171546.

2. Slyvka Y, Inman SR, Malgor R, et al. Protective effects of antioxidant-fortified diet on renal function and metabolic profile in obese Zucker rat. *Endocrine*. 2009;35(1):89–100.

3. Kasiske BL, O'Donnell MP, Keane WF. The Zucker rat model of obesity, insulin resistance, hyperlipidemia, and renal injury. *Hypertension.* 1992;19(1 Suppl):110–115.

4. Blouet C, Mariotti F, Mathe V, et al. Nitric oxide bioavailability and not production is first altered during the onset of insulin resistance in sucrose-fed rats. *Exp Biol Med (Maywood)*. 2007;232(11):1458–1464.

5. Buchwalow I, Schnekenburger J, Tiemann K, et al. L-arginine-NO-cGMP signalling pathway in pancreatitis. *Sci Rep*. 2013;3:1899.

6. Bourgoin F, Bachelard H, Badeau M, et al. Endothelial and vascular dysfunctions and insulin resistance in rats fed a high-fat, high-sucrose diet. *Am J Physiol Heart Circ Physiol*. 2008;295(3):H1044–H1055.

7. Shaker O, Ghallab NA, Hamdy E, et al. Inducible nitric oxide synthase (iNOS) in gingival tissues of chronic periodontitis with and without diabetes: immunohistochemistry and RT-PCR study. *Arch Oral Biol*. 2013;58(10):1397–1406.

8. Schlaich MP, Schmitt D, Ott C, et al. Basal nitric oxide synthase activity is a major determinant of glomerular haemodynamics in humans. *J Hypertens*. 2008;26(1):110–116.

9. Lorin J, Zeller M, Guilland JC, et al. Arginine and nitric oxide synthase: regulatory mechanisms and cardiovascular aspects. *Mol Nutr Food Res*. 2014;58(1):101–116.

10. Luiking YC, Ten Have GA, Wolfe RR, et al. Arginine de novo and nitric oxide production in disease states. *Am J Physiol Endocrinol Metab.* 2012;303(10):E1177–E1189.

11. Jablecka A, Bogdanski P, Balcer N, et al. The effect of oral L-arginine supplementation on fasting glucose, HbA1c, nitric oxide and total antioxidant status in diabetic patients with atherosclerotic peripheral arterial disease of lower extremities. *Eur Rev Med Pharmacol Sci.* 2012;16(3):342–350.

12. Coimbra TM, Janssen U, Grone HJ, et al. Early events leading to renal injury in obese Zucker (fatty) rats with type II diabetes. *Kidney Int.* 2000;57(1):167–182.

13. Toba H, Sawai N, Morishita M, et al. Chronic treatment with recombinant human erythropoietin exerts renoprotective effects beyond hematopoiesis in streptozotocin-induced diabetic rat. *Eur J Pharmacol*. 2009;612(1–3):106–114.

14. Tunctan B, Korkmaz B, Sari AN, et al. Contribution of iNOS/sGC/PKG pathway, COX-2, CYP4A1, and gp91(phox) to the protective effect of 5,14-HEDGE, a 20-HETE mimetic, against vasodilation, hypotension, tachycardia, and inflammation in a rat model of septic shock. *Nitric oxide*. 2013;33:18–41.

15. Prabhakar S, Starnes J, Shi S, et al. Diabetic nephropathy is associated with oxidative stress and decreased renal nitric oxide production. *J Am Soc Nephrol*. 2007;18(11):2945–2952.

16. Kaneki M, Shimizu N, Yamada D, et al. Nitrosative stress and pathogenesis of insulin resistance. *Antioxidants & redox signaling*. 2007;9(3):319–329.

17. Opara EC. Role of oxidative stress in the etiology of type 2 diabetes and the effect of antioxidant supplementation on glycemic control. J Investig Med. 2004;52(1):19–23.

18. Tomohiro T, Kumai T, Sato T, et al. Hypertension aggravates glomerular dysfunction with oxidative stress in a rat model of diabetic nephropathy. *Life Sci.* 2007;80(15):1364–1372.

19. Vega-Lopez S, Devaraj S, Jialal I. Oxidative stress and antioxidant supplementation in the management of diabetic cardiovascular disease. *J Investig Med.* 2004;52(1):24–32.

20. Mekinova D, Chorvathova V, Volkovova K, et al. Effect of intake of exogenous vitamins C, E and beta-carotene on the antioxidative status in kidneys of rats with streptozotocin-induced diabetes. *Nahrung*. 1995;39(4):257–261.

21. Koo JR, Vaziri ND. Effects of diabetes, insulin and antioxidants on NO synthase abundance and NO interaction with reactive oxygen species. *Kidney Int.* 2003;63(1):195–201.

22. Hamilton SJ, Chew GT, Watts GF. Therapeutic regulation of endothelial dysfunction in type 2 diabetes mellitus. *Diab Vasc Dis Res.* 2007;4(2):89–102.

23. Preuss HG. Effects of glucose/insulin perturbations on aging and chronic disorders of aging: the evidence. *J Am Coll Nutr.* 1997;16(5):397–403.

24. Effect of intensive therapy on the development and progression of diabetic nephropathy in the Diabetes Control and Complications Trial. The Diabetes Control and Complications (DCCT) Research Group. *Kidney international*. 1995;47(6):1703–1720.

25. Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). UK Prospective Diabetes Study (UKPDS) Group. *Lancet*. 1998;352(9131):837–853.

26. Coca SG, Ismail-Beigi F, Haq N, et al. Role of intensive glucose control in development of renal end points in type 2 diabetes mellitus: systematic review and meta-analysis intensive glucose control in type 2 diabetes. *Arch Intern Med*. 2012;172(10):761–769.

Citation: Sharon IR, Sonja PM. Investigating inexpensive treatments altering the nitric oxide system for obesity-related type 2 diabetes and diabetic nephropathy. *MOJ Anat Physiol.* 2016;2(2):28–30. DOI: 10.15406/mojap.2016.02.00037