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Dietary Nitrate Lowers Fasting Blood Sugar, HbA1c, Blood Pressure, Improves Sleep in an Elderly - A Case Study

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ABSTRACT
Diabetes Type 2 (T2D) is a pandemic affecting the population the world over. It shows symptoms like hyperglycemia (high blood sugar) in the blood. The insulin hormone is produced by pancreas and insulin resistance is exhibited in T2D. Mitochondrial dysfunction is associated with T2D and generally it is considered as an effect of T2D. Dietary nitrate improves efficiency of mitochondria and we wanted to see if dietary nitrate has any effect on the reduction of symptoms associated with T2D.
In the present case study, we want to report that dietary nitrate has effects on the reduction of symptoms associated with T2D. Moreover, nitrate>nitrite> nitric oxide pathway seen to have its effects on reducing the blood pressure.

I. INTRODUCTION

Diabetics is probably the oldest disease known to man and referred as black-death from the 14th century [1]. Diabetes is prevalent in every population in the world, including rural parts of low- and middle-income countries [2,3]. It is reported, "Just under half a billion people are living with diabetes worldwide and the number is projected to increase by 25% in 2030 and 51% in 2045" [4, 5].

A brief history of the disease, its burden in India and globally, the various methods of management available, is discussed elsewhere [6,7].

The observation of diabetic symptoms in the experimental dogs after removing the pancreas gave an indication of the role of pancreas in diabetics [8]. Subsequently, the discovery of the role of insulin in controlling diabetic in experimental animals [9] confirmed the association of insulin and was well accepted among clinicians until today.

Diabetes in the general population is classified as Diabetes mellitus Type 1 (T1D) and Diabetes mellitus Type 2 (T2D) [10]. Of these, T2D is a major disease among the population all over the world. It is considered as a lifestyle disease caused because of factors like intake of high-calorie diet, lack of exercises along with genetic predisposing factors [11]. Earlier, it was considered as a disease of the affluent. However, its incidences in other countries, especially in Low-Income Countries (LIC) and Middle-Income Countries (MIC) showed that there are other risk factors responsible for the disease [12]. Our observations also support the above studies. In Kerala, a state in South India, there is a high prevalence of the disease not only among top income groups but also among the low and middle income groups who struggle to make two square meals in a day. This led us to look into other probable factors contributing to or triggering the disease.

The entry of glucose into the cells is mediated through the insulin hormone. In T2D condition, the entry of glucose into the cells is slowed down or inhibited even with presence of adequate levels of insulin in the blood. This condition is called insulin resistance, where insulin becomes inactive in transporting the glucose effectively into the cells because of some unknown reasons. This leads to building up of glucose level in the blood and this condition is termed as hyperglycemia. Prolonged hyperglycemic conditions may affect other organs and cells, demanding removal of glucose from the blood through pharmacological interventions.

The reason for insulin resistance is not clearly understood until now. Many publications have reported an association between mitochondrial dysfunction with T2D.

Nitrates were considered as a foe in human nutrition because of its association with certain cancers. But now it has gained importance in human nutrition in recent times, especially after finding the importance of the Nitrate (NO3) > Nitrite (NO2) > Nitric oxide (NO) pathway [13,14].

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As stated above, there is an association between mitochondrial dysfunction and T2D. Recently, it is reported that dietary nitrate improves the efficiency of human mitochondria [15,16]. With this information in the background, we wanted to test whether dietary nitrate has any effect on lowering Fasting Blood Sugar (FBS) and HbA1C in a diabetic patient, probably by improving the mitochondrial function.

II. CASE STUDY

Patient's background

A 65-year-old elderly male with a history of T2D over 10 years, hypertensive, had two episodes of ccongestive heart failure (CHF) in the past and with low LV function (eff <20) wanted to test whether dietary nitrate has any effect in lowering the blood sugar and HbA1C.

The patient used to control his T2D by restricting his calorie intake and walking at least 2 Km in a day. Thus, he was never on any medication. This routine was upset in March 2020 when the patient had an episode of CHF followed by covid-19 lockdown, restricting his movements. In May 2020, the patient had shingles and in the first week of August 2020; the patient had COVID-19. The patient recovered from all the situations. However, the patient had general weakness, apnea and difficulty in breathing while lying down. Thus, the patient was admitted and treated for four days in a hospital. He was on medications for hypertension and diabetes. He was also on diuretics. The discharge summary is given in Table 1 in the results section.

Life style adopted by the patient during the study

The patient, after getting discharged from the hospital, made some changes in his lifestyle.

1) The patient had a general knowledge of nitrate metabolism in the body and understood the importance of micro flora in the mouth in converting nitrate to nitrites. So he avoided conventional tooth pastes for flossing. Patient chose "umikkari" (activated charcoal made of rice husks) for cleaning the teeth with a tooth brush.

2) Every morning after getting up, the patient drank half a glass of water to consume the nitrite rich saliva. This facilitates the conversion of nitrite to nitric oxide in the presence of acids in the stomach.

3) The patient avoided water filtered through Reverse Osmosis (RO) as RO removes nitrates in the drinking water and It was replaced with 1.5 liters of water supplied by the public distribution system, which is sourced from reservoirs and rivers.

4) Food taken during dinner was experimented on to see which diet gave lower fasting blood sugar levels (FBS) the next morning..

5) Dietary nitrate was incorporated as spinach soup or other cuisine rich in leafy vegetables. The procedure used for making spinach soup is given elsewhere [17].

6) Patient avoided all processed food including table salt with anti-caking agents and relied only on food prepared at home using raw sea salt. The study was carried out for about a year, and the observations were recorded in a personal file.

7) The patient used moderate free sugar for his two or three cups of tea in a day.

8) Patient continued walking at least 2Km in a day, throughout the study.

III. RESULTS

The levels of FBS and HbA1C at the time of discharge from the hospital are given in Table 1.

The levels of FBS at different time periods are given in Table 2.

A. **Quantitative analysis of parameters (fasting blood sugar and HbA1C with respect to dietary nitrate.**

Date	Fasting Blood Sugar (FBS) mg/dl	HbA1C	Lifestyle followed
26th October, 2020	182	8.7	
2nd November, 2020	167		Started the lifestyle changes mentioned above

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11th November, 2020	165		
21 November, 2020	139		
23rd November,2020	123		
25 November,2020	123		
9th January, 2021	139		
26th January, 2021	121		
10th February, 2021	121		
26th February,2021	121		

Table 1. Fasting Blood Sugar (FBS) mg/dl) and HbA1C level in the patient at the time of discharge from the hospital.

Date	Fasting Blood Sugar (FBS) mg/dl		Lifestyle followed
28th August, 2021	125		
			Changed to an ordinary diet with whatever is available.
1st January,2022	127		

Table 2. The fasting blood sugar (FBS) mg/dl and the HbA1C of the patient after changing the lifestyle.

B. **Qualitative description by the patient on the effect of dietary nitrate**

The results presented in the Table 1 and 2 shows that dietary nitrate influenced the health status of the patient and it is self explanatory.

Flossing with activated charcoal did not create any foul smell in the mouth or breath.

There was marked improvement in sleep and general weakness. There was improvement in erectile dysfunction associated with T2D.

IV. DISCUSSION

Diabetes is a metabolic disorder where the levels of glucose rises in the blood (hyperglycemia) even with adequate levels of insulin in the blood

[18,19] Hyperglycemia resulting from the insulin resistance is the first symptom of T2D and the reasons for insulin resistance is not known [20].

Glucose, a major source of fuel, enters inside the cell and it is used for energy production through chemical breakdown of the glucose molecule and through electron transfer through certain chemicals. Finally, oxygen acts as the final electron acceptor and accepts the electrons to form water molecules. This takes place in the electron transport chain in the mitochondria and the mitochondria are considered as the powerhouse of cells.

Mitochondrial dysfunction is reported to be associated with T2D, and it is considered as an effect of T2D [21]. Thus, the medical fraternity was targeting to tackle insulin resistance to combat hyperglycemia [22].

The present treatment modalities include suggesting food with low glycemic index so that the glucose available in the intestine after digestion may be reduced. Another method is to remove excess glucose in the blood through kidneys using some drugs, just like jettisoning extra fuel from aircraft before landing, to achieve its prescribed landing weight. The third method is to use drugs like metformin that will influence the enzyme adenosine monophosphate kinase (AMPK) which is involved in the inhibition of enzymes involved in stimulating insulin signaling and glucose transport in muscles, gluconeogenesis and glycogen synthesis [23], AMPK regulates the cellular and organs [23].

Against such a backdrop, based on our previous work on the study of physiological and biochemical responses of *A. granosa* to naphthalene, we suggested that insulin resistance could be a feedback mechanism [24]. Therefore, we proposed that mitochondrial dysfunction could be a triggering factor in T2D [24].

Mitochondrial dysfunction could arise because of various factors, as discussed elsewhere [25]. The reduction of diabetic symptoms observed in the patient taking home cooked food with no preservatives, support our hypothesis. Many processed foods use preservatives to increase its shelf life. The preservative levels may be well below the Allowable Daily Intake (ADI) prescribed. However, when food with preservatives is taken regularly without giving enough time for its clearance from the body, it may build its concentration in the body with time. Moreover, there is a lack of information on the effect of low concentrations of preservatives on the vital organs like mitochondria. In the present study, home cooked food with no preservatives may have helped better functioning of mitochondria.

Nitrate was primarily used as a preservative in meat processing industries [26] and when the meat was cooked by heat produced nitrosamines [27], a carcinogenic compound. This caused an increase in cancer incidences in the population consuming preserved meat products. Thus, nitrate was considered as a foe to human nutrition and maximum allowable nitrate in the drinking water was also suggested [28].

Recent studies have shown the importance of Nitrate > Nitrite > Nitric oxide pathway in human nutrition [13 - 16] as discussed earlier.It is estimated that an adult requires about 3.7 mg/kg BW/day or at least 222 mg of nitrate daily for a person with 60 Kg [29,30]. About 10mg/l of nitrate, in drinking water, is considered as safe limits [31]. But with the Reverse Osmosis to remove the hardness removes nitrates along with other elements [32]. Thus, the levels of nitrates in the drinking water obtained by reverse osmosis are very low or nil. This is the reason for the patient depending on the drinking water from the public water supply system. The public water supply delivers filtered water from the reservoirs and rivers without removing and nitrates present in them.

In the present study, nitrate was supplied from the spinach soup. Spinach contains an exorbitant amount of nitrate and this may have supplied adequate amounts of nitrate [29]. The dietary nitrate and metformin behave in the same manner in activating the AMPK pathway [13-14]. The dietary nitrate also improves the functioning of mitochondria [15]. Thus, all these factors may have contributed to the improvement of mitochondria and glucose utilization, leading to lower glucose level in the blood.

The BP of the patient was always stable and within the normal limits. In the nitrate > nitrite > nitric oxide pathway, the contribution of microflora in the mouth is important. The usual toothpaste contains anti-bacterial chemicals which may hamper the pathway. Drinking of water in the morning helped to save nitrite formed in the mouth

and transported to stomach where it may have been converted to nitric oxide in the presence of acids. The increased nitric oxide may have helped in controlling the BP at normal levels. Nitric oxide helps in the function of endothelium of arteries and capillaries. Its effect was noticeable with a reduction in the erectile dysfunction observed with dietary intake of nitrate and using activated charcoal for flossing the teeth. Incidentally, some communities in rural Maharashtra practice the drinking water as soon as they get up to control diabetes [33]. They did not know the reason for this practice but definitely saw the result. This once again suggests that intuition leads man for his development [34]..

In the present case study, HbA1C was lowered to. 6.6 from 8.7. In our earlier study using wheat porridge in the diet, the HbA1C was about 5.7 [35]. We did not use wheat porridge in the present study because we wanted to evaluate the role of dietary nitrate in lowering hyperglycemia.

The present study has many limitations. But it throws some light on the importance of dietary nitrate in improving the functioning of mitochondria, provided there is a reduced intake of electron chain inhibitors found in food preservatives. Thus, we want to propose that inclusion of wheat porridge and dietary nitrate may help the patient to control hyperglycemia in T2D patients. We have seen many patients who could not undergo surgery because of hyperglycemia benefitted with intake of wheat porridge for a week or two in bringing down the sugar level. The activated charcoal to clean teeth seems to improve nitric oxide production in the body, evidenced by stable BP and reduced erectile dysfunction.

We also want to conclude that the present study supports our hypothesis that mitochondrial dysfunction caused by various electron transport inhibitors may slow down glucose utilization in the cells. The insulin resistance observed in T2D is a feedback mechanism that prevents uncontrolled entry of glucose into the cellular cytoplasm because uncontrolled glucose entry may demand complicated Osmo-regulation with additional energy expenditure.

The present study supports the hypothesis “if nitrate in green vegetables or given as a salt via a dietary supplement is protective, this can rapidly be translated into cost-efficient and highly specific dietary approaches to prevent the development of disease or to treat patients with T2D" [14].

V. CONCLUSION

The present case study supports that mitochondria is the primary organ affected by various electron transport inhibitors in the diet which slows down utilization of glucose molecules. Uncontrolled entry of glucose into the cells demands intricate osmo-regulation with the demand of water molecules. Water is very scarce resource for terrestrial animals and hence insulin may act as a gatekeeper that controls the entry of glucose into the cells. Thus hyperglycemia observed in T2D could be the result of mitochondrial dysfunction. The present case study shows the efficacy of dietary nitrate in improving the functioning of mitochondria that led to better utilization of blood glucose, supporting the hypothesis that mitochondrial dysfunction could be the primary event associated with T2D.

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Conflict of Interest

Nil

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REFERENCES

[1]. Deepthi B, Sowjanya K, Lidiya B, et al. A Modern Review of Diabetes Mellitus: An Annihilatory Metabolic Disorder. J In Silico In Vitro Pharmacol. 2017, 3:1

[2]. Editors. https://www.who.int/news-room/fact-sheets/detail/diabetes

[3]. Lin X, Xu Y, Pan X, Xu J, Ding Y, Sun X, Song X, Ren Y, Shan PF. Global, regional, and national burden and trend of diabetes in 195 countries and territories: an analysis from 1990 to 2025. Sci Rep. 2020 Sep 8;10(1):14790. doi: 10.1038/s41598-020-71908-9. PMID: 32901098; PMCID: PMC7478957.

[4]. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, Colagiuri S, Guariguata L, Motala AA, Ogurtsova K, Shaw JE, Bright D, Williams R; IDF Diabetes Atlas Committee. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045:

Results from the International Diabetes Federation Diabetes Atlas, 9th edition. Diabetes Res Clin Pract. 2019 Nov;157:107843. doi: 10.1016/j.diabres.2019.107843. Epub 2019 Sep 10. PMID: 31518657.

[5]. Diabetes Atlas Committee. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the International Diabetes Federation Diabetes Atlas, 9th edition. Diabetes Res Clin Pract. 2019 Nov;157:107843. doi: 10.1016/j.diabres.2019.107843. Epub 2019 Sep 10. PMID: 31518657.

[6]. Latha S , Vijayakumar R. The Facts about Diabetes Mellitus- A Review. Galore International Journal of Health Sciences and Research (www.gijhsr.com) 71 Vol.4; Issue: 2; April-June 2019

[7]. Vijayakumar, G., Manghat, S., Vijayakumar, R. et al. Incidence of type 2 diabetes mellitus and prediabetes in Kerala, India: results from a 10-year prospective cohort. BMC Public Health **19,** 140 (2019). https://doi.org/10.1186/s12889-019-6445-6

[8]. Dittrich HM, Hahn von Dorsche H. Die anatomisch-histologische Erforschung des Pankreas vom Beginn des 19. Jahrhunderts bis zur Entdeckung des insulins durch Banting und Best (1921) unter Berücksichtigung physiologischer Aspekte. Die Entdeckung des Pankreasdiabetes durch von Mering und Minkowski (1889) [The anatomical and histological investigation of the pancreas in the 19th century and till the discovery of insulin (1921) 3. The discovery of the pancreas-diabetes by von Mering and Minkowski (1889) (author's transl)]. Anat Anz. 1978;143(5):509-17. German. PMID: 358862.

[9]. Banting FG Best CH Collip JB Hepburn J Macleod JJR Noble EC. Preliminary studies of the physiological of insulin. Trans R Soc Can. (1922) 16:18.

[10]. Editors. Classification of diabetes mellitus. Geneva: World Health Organization; 2019. License: CC BY-NC-SA 3.0 IGO.

[11]. Editors. Diseases of affluence. https://en.wikipedia.org/wiki/Diseases_of_affluence Accessed on 14th November, 2021.

[12]. Dagenais GR, Gerstein HC, Zhang X, McQueen M, Lear S, Lopez-Jaramillo P, Mohan V, Mony P, Gupta R, Kutty VR, Kumar R, Rahman O, Yusoff K, Zatonska K, Oguz A, Rosengren A, Kelishadi R, Yusufali A, Diaz R, Avezum A, Lanas F, Kruger A, Peer N, Chifamba J, Iqbal R, Ismail N, Xiulin B, Jiankang L, Wenqing D, Gejie Y, Rangarajan S, Teo K, Yusuf S. Variations in Diabetes Prevalence in Low-, Middle-, and High-Income Countries: Results From the Prospective Urban and Rural Epidemiological Study. Diabetes Care. 2016 May;39(5):780-7. doi: 10.2337/dc15-2338. Epub 2016 Mar 10. PMID: 26965719.

[13]. Lundberg, J., Gladwin, M., Ahluwalia, A. et al. Nitrate and nitrite in biology, nutrition and therapeutics. Nat Chem Biol **5,** 865–869 (2009). https://doi.org/10.1038/nchembio.260

[14]. Lundberg JO, Carlström M, Weitzberg E. Metabolic Effects of Dietary Nitrate in Health and Disease. Cell Metab. 2018 Jul 3;28(1):9-22. doi: 10.1016/j.cmet.2018.06.007. PMID: 29972800.

[15]. Larsen FJ, Schiffer TA, Borniquel S, Sahlin K, Ekblom B, Lundberg JO, Weitzberg E. Dietary inorganic nitrate improves mitochondrial efficiency in humans. Cell Metab. 2011 Feb 2;13(2):149-59. doi: 10.1016/j.cmet.2011.01.004. PMID: 21284982.

[16]. Nair KS, Irving BA, Lanza IR. Can dietary nitrates enhance the efficiency of mitochondria? Cell Metab. 2011 Feb 2;13(2):117-8. doi: 10.1016/j.cmet.2011.01.013. PMID: 21284976; PMCID: PMC3049330.

[17]. Editor. Best bite.Healthy Spinach Soup. Available in https://www.youtube.com/watch?v=f9UoLQFFgWk

[18]. Hameed I, Masoodi SR, Mir SA, Nabi M, Ghazanfar K, Ganai BA. Type 2 diabetes mellitus: From a metabolic disorder to an inflammatory condition. World J Diabetes. 2015 May 15;6(4):598-612. doi: 10.4239/wjd.v6.i4.598. PMID: 25987957; PMCID: PMC4434080.

[19]. Freeman AM, Pennings N. Insulin Resistance. [Updated 2022 Jul 4]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK507839/

[20]. Higuera V. What is Insulin resistance? Causes, symptoms, diagnosis, treatment and Prevention. Available at https://www.everydayhealth.com/type-2-

diabetes/insulin-resistance-causes-symptoms-diagnosis-consequences/

[21]. Lowell BB, Shulman GI. Mitochondrial dysfunction and type 2 diabetes. Science. 2005 Jan 21;307(5708):384-7. doi: 10.1126/science.1104343. PMID: 15662004.

[22]. Saini V. Molecular mechanisms of insulin resistance in type 2 diabetes mellitus. World J Diabetes. 2010 Jul 15;1(3):68-75. doi: 10.4239/wjd.v1.i3.68. PMID: 21537430; PMCID: PMC3083885.

[23]. Nasri H, Rafieian-Kopaei M. Metformin: Current knowledge. J Res Med Sci. 2014 Jul;19(7):658-64. PMID: 25364368; PMCID: PMC4214027.

[24]. Eapen, JTl. Insulin resistance: is it a feedback mechanism or a therapeutic target in type 2 diabetes?. **International Journal of Research in Medical Sciences**, [S.l.], v. 7, n. 8, p. 3202-3205, july 2019. ISSN 2320-6012. Available at: <https://www.msjonline.org/index.php/ijrms/article/view/6685>. Date accessed: 07 aug. 2022. doi:http://dx.doi.org/10.18203/2320-6012.ijrms20193421.

[25]. Pizzorno J. Mitochondria-Fundamental to Life and Health. Integr Med (Encinitas). 2014 Apr;13(2):8-15. PMID: 26770084; PMCID: PMC4684129.

[26]. Ferysiuk K, Wójciak KM. Reduction of Nitrite in Meat Products through the Application of Various Plant-Based Ingredients. Antioxidants (Basel). 2020 Aug 5;9(8):711. doi: 10.3390/antiox9080711. PMID: 32764511; PMCID: PMC7464959.

[27]. Scanlan RA. Formation and occurrence of nitrosamines in food. Cancer Res. 1983 May;43(5 Suppl):2435s-2440s. PMID: 6831466.

[28]. Ward MH, Jones RR, Brender JD, de Kok TM, Weyer PJ, Nolan BT, Villanueva CM, van Breda SG. Drinking Water Nitrate and Human Health: An Updated Review. Int J Environ Res Public Health. 2018 Jul 23;15(7):1557. doi: 10.3390/ijerph15071557. PMID: 30041450; PMCID: PMC6068531.

[29]. European Food Safety Authority. Nitrate in vegetables EFSA Journal. 2008;68:91–79.

[30]. Brkić D, Bošnir J, Bevardi M, Bošković AG, Miloš S, Lasić D, Krivohlavek A, Racz A, Ćuić AM, Trstenjak NU. NITRATE IN LEAFY GREEN VEGETABLES AND ESTIMATED INTAKE. Afr J Tradit Complement Altern Med. 2017 Mar 1;14(3):31-41. doi: 10.21010/ajtcam.v14i3.4. PMID: 28480414; PMCID: PMC5412236.

[31]. Ward MH, Jones RR, Brender JD, de Kok TM, Weyer PJ, Nolan BT, Villanueva CM, van Breda SG. Drinking Water Nitrate and Human Health: An Updated Review. Int J Environ Res Public Health. 2018 Jul 23;15(7):1557. doi: 10.3390/ijerph15071557. PMID: 30041450; PMCID: PMC6068531

[32]. Shrimali M, Singh KP. New methods of nitrate removal from water. Environ Pollut. 2001;112(3):351-9. doi: 10.1016/s0269-7491(00)00147-0. PMID: 11291441.

[33]. Abraham KM. Administrator, Union Biblical Seminary, Yeotmal, Maharashtra. (Personal communication (1980).

[34]. Eapen JT. Mankind's amazing progress- Is it based on science and technology or intuition from God? International Journal of Creative Research Thoughts (IJCRT) 6(2): 181-189.

[35]. **E**apen JT., "Let food be your medicine.." Inclusion of wheat porridge, a whole grain meal, and legumes lowers postprandial glucose in diabetic patients. Journal of Medical Research and Innovation 1(2), https://doi.org/10.5281 \