Research Article
Volume 15:01, 2023

Journal of Bioanalysis & Biomedicine

ISSN: 1948-593X **Open Access**

Antioxidant Trace Elements Depletion Induces Proinflammatory Cytokines Polarization in Anti Diabetics Drug Naive Non Obese Male Type 2 Diabetic Nigerians

Ehiaghe Friday Alfred*[1], Udiyiwe Osaikhuwuomwan[2], Uwaifo Nicholas Eghosa[3], Ehiaghe Imuetinyan Joy[4], Eremwanarue Aibuedefe Osagie[5], Usiosefe Raphael Erhunmwunse[4], Digban Awharentomah Kester[6] and Emeje Paul Isaac[7]

[1]Department of Medical Laboratory Science, Nnamdi Azikiwe University, Awka, Nigeria
[2]Department of Medical Laboratory Science, Igbinedion University, Okada, Edo State, Nigeria
[3]Department of Medical Laboratory Science, Igbinedion University Teaching Hospital, Okada, Edo State, Nigeria
[4]Department of Medical Laboratory Science, Benson Idahosa University, Benin City, Edo State Nigeria
[5]Department of Plant Biology and Biotechnology, University of Benin, Edo State, Nigeria
[6]Department of Medical Laboratory Science, Novena University Delta State, Nigeria
[7]Department of Medical Laboratory Science, Kogi State University, Anyigba

Abstract

Type 2 diabetes (T2D) is a major risk factor for cardiovascular diseases and acute oxidative stress (OS) by high production of reactive oxygen species (ROS) related to the lipotoxicity and glucotoxicity processes. The study was aimed at evaluating the cytokine responses and antioxidant trace elements status using the blood levels of interleukin-1 alpha, interleukin-4, zinc nickel, magnesium, and chromium circulating among anti-diabetics drug naïve non-obese male type 2 Diabetic Nigerians. A total of 124 subjects (aged between 20 and 40 year) were randomly studied, among these were 64 known cases of type 2 diabetes mellitus with a poor glycemic control index and 60 apparently healthy individuals with a good glycemic index. Student's t- test was used to compare independent variables. Cytokine levels were determined by enzyme-linked immunosorbent assay method. Heavy metals were analyzed by atomic absorption spectrophotometer method. Full blood count was done using haematology auto-analyzer. The probability values less than 0.05 were considered significant. The levels of interleukin -1α alpha were significantly higher in the diabetics group as compared with the control group ($p<0.002$), whereas, The levels of interleukin-4, packed cell volume, total white blood count, red blood cells, mean cell haemoglobin and mean cell haemoglobin concentration were significantly lower as compared with the control group ($P<0.005$). The levels of Manganese, Chromium, Nickel, Zinc were significantly lower in the diabetics group as compared with the control group ($p<0.002$). Depletion of antioxidant trace elements in T2D is linked to increased pro-inflammatory cytokines and may contribute to the development of diabetic complications. ATE supplementation may be able to prevent the observed cytokine imbalance.

Keywords: Cytokine responses • Type 2 diabetes mellitus • Cytokine balance • Antioxidant trace

Introduction

T2D is a substantial risk factor for cardiovascular disease and acute oxidative stress (OS) due to the increased generation of reactive oxygen species (ROS) associated with the lipotoxicity and glucotoxicity processes [1]. It is not entirely apparent how cytokines responses and the level of antioxidant trace elements (ATE) such nickel (Ni), zinc (Zn), magnesium (Mg), and chromium (Cr) affect immunological diseases [2]. However, the function of ATE as an important micronutrient has long been recognized as a possible option for treating metabolic diseases, such as glucose homeostasis in prediabetes [3]. Several studies have reported that the pathogenesis of type 2 diabetes (T2D) is related to an imbalance of some antioxidant trace elements (zinc, selenium, copper, manganese, chromium), which may adversely affect pancreatic islets and cause diabetes development, and is clearly associated with ROS production and insulin signaling [4]. T2D is related with elevated pro-inflammatory cytokines (TNF-, IL-6) that may contribute to the development of diabetes complications [5-7] and increased glycated haemoglobin production [4]. Several studies have shown that type 2 diabetes (T2D) is a low-grade inflammatory condition, and that lowering inflammation is a key result for T2D therapy because it causes a balance between pro-inflammatory and anti-inflammatory cytokines [8-12]. The toxicological effects of unregulated reactive oxygen species production have been linked to various metabolic defects such as cardio-vascular attacks and auto-immune disorders, but there is still little known about the cytokine responses of anti-diabetics drug naive non-obese male type 2 diabetics in Nigeria. To the best of our knowledge, this is the first research to assess cytokine responses and antioxidant trace element status utilizing blood levels of interleukin-1 alpha, interleukin-4, zinc nickel, magnesium, and chromium circulating among anti-diabetics drug naive non-obese male type 2 diabetic Nigerians.

***Address for Correspondence:** Ehiaghe Friday Alfred, Department of Medical Laboratory Science, Nnamdi Azikiwe University, Awka, Nigeria, E-mail: fa.ehiaghe@unizik.edu.ng*

Received: 19 December 2022, Manuscript No. jbabm-22-84896; **Editor assigned:** 21 December, 2022, PreQC No. P-84896; **Reviewed:** 04 January 2023, QC No. Q-84896; **Revised:** 09 January 2023, Manuscript No. R-84896; **Published:** 16 January, 2023, DOI: 10.37421/1948-593X.2022.15.364

Materials and Methods

Before the commencement of the case-control study, ethical approvals were obtained from UBTH Ethical Review Committee. A total of 124 subjects (aged between 20 and 40 year) were randomly studied, among these were 64 known cases of type II diabetes mellitus with a poor glycemic control index and 60 apparently healthy individuals with a good glycemic index (see table 1). Informed consent was obtained from all subjects before the commencement of the study.

Collection of sample: For the heavy metals determination, about 3 ml of venous blood samples were collected aseptically into Potassium ethylene diamine tetra acetic acid (K-EDTA) anticoagulant tubes. The samples were

labelled and immediately placed in ice pack at the site of collection and subsequently transferred into a refrigerator at 4ºC. For the full blood count (FBC) and Interleukin assays, 4 ml of venous blood were collected into K-EDTA tubes, mixed and FBC carried out immediately. The blood samples were centrifuged within 30 minutes of collection for 15 minutes at 2000 rounds per minute (RPM) and plasma separated from the cells into plain tubes and refrigerated at 2-8ºC.

Determination of blood zinc, magnesium, chromium and nickel and cytokines: A calibrated Buck 205 flame atomic absorption spectrophotometer (Perkin-Elmer, HGA-2100) was used to analyze the blood sample for Zinc, Magnesium, Chromium and Nickel as described by Iyengar GV, et al. [13] at Yitzhak Rabin Laboratory for Advanced Micropropagation and Biotechnology Research Centre, Nnamdi Azikiwe University Awka, Nigeria. This method is based on the principle that atoms of the element when aspirated into Atomic Absorption Spectrophotometer vaporizes and absorbed light of the same wavelength as that emitted by the element when in the excited state. The amount of light absorbed can be correlated in a linear fashion to the concentration of the analyte in the sample. The levels of IL-1β and IL-4 were determined using enzyme-linked immunosorbent assay (ELISA) method as described by Alfred EF, et al. [14].

Data analysis: Student's t- test was used to compare independent variables. The probability values less than 0.05 were considered significant. The statistical analysis were done using SPSS version 20.0.

Results

(Table 1). Shows the mean ± (SD) of age and body mass index of the studied non obese diabetics group with the control group (Table 2). Shows the mean ± (SD) Levels of interleukin -1α, interleukin-4 and some haematological parameters of diabetics group and control group. The levels of interleukin -1α were significantly higher in the diabetics group as compared with the control group ($p<0.002$), whereas, The levels of interleukin-4, packed cell volume, total white blood count, red blood cells, mean cell haemoglobin and mean cell haemoglobin concentration were significantly lower as compared with the control group ($P<0.005$) (Table 3). Shows the mean ± (SD) levels of antioxidant trace elements in diabetics group as compared with the control group. The levels of Manganese, Chromium, Nickel, Zinc were significantly lower the diabetics group as compared with the control group ($p<0.002$).

Table 1. Statistical analysis of glycated hemoglobin, age and body mass index values in non obese diabetics group and control group (mean ± SD).

Parameters	Non-obese diabetics group (n= 64)	Control group (N = 60)
HbA1c (%)	9.60 ± 0.67*	5.9 ± 0.06
Age (years)	36.9 ± 3.17	37.5 ± 2.08
BMI (kg/m^2)	27.5 ± 1.05*	19.6 ± 1.03

Table 2. Statistical analysis of interleukin-4, interleukin -1α, and some haematological values in non-obese diabetics group and control group (mean ±SD).

Parameters	Non-obese Diabetics group	Control group	t- value	p-value
Interleukin-4 (ng/L)	10.1 ± 0.45*	14.3 ± 0.08	2.354	0.026
Interleukin-1α (ng/L)	97.6 ± 0.62*	16.4 ± 1.10	-2.481	0.023
WBC (10^3/µL)	3.8 ± 0.32*	5.7 ± 0.42	0.034	0.042
LYM (%)	45.7 ± 2.83	45.1 ± 2.36	0.157	0.876
MID (%)	4.5 ± 0.82	4.8 ± 0.46	4.059	0.974
GRAM (%)	40.8 ± 3.83	50.2 ± 2.69	-1.958	0.058
RBC (10^6/µL)	4.8 ± 0.12*	5.2 ± 0.15	-2.099	0.042
HGB (g/dL)	10.9 ± 0.35*	13.8 ± 0.15	0.179	0.859
HCT (%)	30.7 ± 1.04*	41.6 ± 1.02	-2.404	0.021
MCV (fl)	81.8 ± 1.23	83.0 ± 1.20	-0.688	0.496
MCH (pg)	25.1 ± 0.46*	26.9 ± 0.64	-2.283	0.028
MCHC (g/dL)	30.6 ± 0.41*	33.0 ± 0.17	-5.234	0.001

Table 3. Statistical analysis of trace element values in non-obese diabetics group and control group (mean ± SD).

Parameters	Non-obese diabetics group	Control group	t-values	P-values
Mg (µg/L)	90.9 ± 1.68*	126.6 ± 1.65	1.850	0.032
Nickel (µg/L)	168.3 ± 7.84*	249.4 ± 8.92	10.048	0.001
Zinc (µg/L)	101.5± 13.28*	192.4± 10.72	- 5.254	0.001
Cr (µg/L)	51.1 ± 11.50*	78.7 ± 14.72	0.128	0.04

Discussion

The observed significant increase in proinflammatory interleukin-1 levels in this study, coupled with significantly lower levels of interleukin-4, packed cell volume, total white blood count, red blood cells, mean cell haemoglobin, and mean cell haemoglobin concentration, could be associated with a depletion of antioxidant trace elements (Manganese, Chromium, Nickel, Zinc), which induces a state of low-grade inflammation (see table 2 & 3). Several researchers have also expressed their views in this regard, however they have emphasized that Zn is an important ATE component because: (i) it plays a vital function in the stability of insulin hexamers and hormone pancreatic storage [15], and (ii) it is an effective antioxidant [16]. Lower Zn plasma concentrations were reported in T2D is a risk factor for cardiovascular-metabolic syndrome [17,18] and lower Zn levels in diabetics appear to be associated to an increased risk for coronary artery disease [19]. It has been proven that ATE-induced immune cell activation contributes to the proinflammatory environment of the diabetic islet [20]. It has been found that higher levels of IL-6, IL-8, TNF-, and IL-1 beta are connected with elevated blood glucose levels seen in T2D patients [21-25] and ATE deficiency results in lower levels of the anti-inflammatory cytokine. The current investigation supports the findings of Jagannathan B, et al. [26], who found that blood from T2D patients with ATE depletion had higher levels of circulating Th17 cells, which are proinflammatory T cell subsets. Reduced inflammation has been proven in studies to be an essential result for T2D therapy because it causes a balance between pro-inflammatory and anti-inflammatory cytokines [27-30]. However, we hypothesize that ATE supplementation may decrease the overproduction of pro-inflammatory cytokines and reactive oxygen species caused by ATE deficiency.

Conclusion

Depletion of antioxidant trace elements in T2D is linked to increased pro-inflammatory cytokines and may contribute to the development of diabetic complications. ATE supplementation may be able to prevent the observed cytokine imbalance.

Competing Interests

The authors declare that they have no competing interests.

Authors' Contributions

This work was carried out in collaboration between all authors. Author EFA and UO designed the study and performed the statistical analysis. Authors EFA, UO, UNE, DAK, EJI, EAO, URE and EPI conducted and managed the Laboratory analysis. All authors read and approved the final manuscript.

Acknowledgement

We acknowledge the cooperation of the members of staff of Iykenson Medicals and Research Centre, Awka, Nigeria.

References

1. Zhang, Jixiang, Xiaoli Wang, Vikash Vikash and Qing Ye, et al. "ROS and ROS-mediated cellular signaling." *Oxid Med Cell Longev* (2016).
2. Andersen, Ole and Jeanne Freeland-Graves. "Recent developments in trace element research." *J Trace Elem Med Biol* 2 (2012): 59-60.
3. Xiu, Y. M. "Trace elements in health and diseases." *Biomed Environ Sci* 9 (1996): 130-136.
4. Wiernsperger, Nicolas and JeanRobert Rapin. "Trace elements in glucometabolic disorders: An update." *Diabetol Metab Syndr* 2 (2010): 1-9.
5. Badran, M., R. Morsy, H. Soliman and T. Elnimr. "Assessment of trace elements levels in patients with type 2 diabetes using multivariate statistical analysis." *J Trace Elem Med Biol* 33 (2016): 114-119.
6. Evans, Joseph L., Betty A. Maddux and Ira D. Goldfine. "The molecular basis for oxidative stress-induced insulin resistance." *Antioxid Redox Signal* 7 (2005): 1040-1052.
7. Van Campenhout, Ann, Christel Van Campenhout, Albert R. Lagrou and Pascale Abrams, et al. "Impact of diabetes mellitus on the relationships between iron inflammatory and oxidative stress status." *Diabetes Metab Res R* 22 (2006): 444-454.
8. Viktorínová, Alena, Eva Tošerová, Marián Križko and Zdenka durackova. "Altered metabolism of copper, zinc, and magnesium is associated with increased levels of glycated hemoglobin in patients with diabetes mellitus." *Metab* 58 (2009): 1477-1482.
9. Uysal, K. Teoman, Sarah M. Wiesbrock, Michael W. Marino and Gkhan S. Hotamisligil. "Protection from obesity-induced insulin resistance in mice lacking TNF-α function." *Nature* 389 (1997): 610-614.
10. Hirosumi, Jiro, Gürol Tuncman, Lufen Chang and Cem Z. Görgün, et al. "A central role for JNK in obesity and insulin resistance." *Nature* 420 (2002): 333-336.
11. Yuan, Minsheng, Nicky Konstantopoulos, Jongsoon Lee and Lone Hansen, et al. "Reversal of obesity-and diet-induced insulin resistance with salicylates or targeted disruption of Ikkβ." *Sci* 293 (2001): 1673-1677.
12. Hundal, Ripudaman S., Kitt F. Petersen, Adam B. Mayerson and Pritpal S. Randhawa, et al. "Mechanism by which high-dose aspirin improves glucose metabolism in type 2 diabetes." *J Clin Investig* 109 (2002): 1321-1326.
13. Iyengar, G. V., Subramanian, K. S and Woittiez, J. R. "Element analysis of biological samples: Principles and practice." *CRC Press* (2020).
14. Alfred, E. F., A. Benson Olu, E. Imuetiyan Joy and J. Sunday, et al. "The levels of c-reactive protein, malondialdehyde and absolute lymphocyte counts in pre and post-acute exercise." *J Sports Med Doping Stud* 7 (2017): 2161-0673.
15. Wijesekara, N., F. Chimienti and M. B. Wheeler. "Zinc, a regulator of islet function and glucose homeostasis." *Diabetes Obes Metab* 11 (2009): 202-214.
16. Prasad, Ananda S. "Clinical, immunological, anti-inflammatory and antioxidant roles of zinc." Experimental gerontology 43 (2008): 370-377.
17. Singh, Ram B., Mohammad A. Niaz, Shanti S. Rastogi and Sarita Bajaj, etal. "Current zinc intake and risk of diabetes and coronary artery disease and factors associated with insulin resistance in rural and urban populations of North India." *J Am Coll Nutr* 17 (1998): 564-570.
18. Obeid, Omar, Manal Elfakhani, Sani Hlais and Michel Iskandar, et al. "Plasma copper, zinc, and selenium levels and correlates with metabolic syndrome components of lebanese adults." *Biol Trace Elem Res* 123 (2008): 58-65.
19. Soinio, Minna, Jukka Marniemi, Markku Laakso and Kalevi Pyorala, Seppo Lehto, et al. "Serum zinc level and coronary heart disease events in patients with type 2 diabetes." *Diabetes care* 30 (2007): 523-528.
20. Bahgat, Mervat and Dalia Ibrahim. "Proinflammatory cytokine polarization in type 2 diabetes." *Eur J Immunol* 45 (2020): 170-175.
21. Mosser, David M and Justin P. Edwards. "Exploring the full spectrum of macrophage activation." *Nat Rev Immunol* 8 (2008): 958-969.
22. Pickup, John C. "Inflammation and activated innate immunity in the pathogenesis of type 2 diabetes." *Diabetes care* 27 (2004): 813-823.
23. Crook, M. "Type 2 diabetes mellitus: A disease of the innate immune system? an update." Diabetic Medicine 21 (2004): 203-207.
24. Giulietti, Annapaula, Evelyne van Etten, Lut Overbergh and Katinka Stoffels, et al. "Monocytes from type 2 diabetic patients have a pro-inflammatory profile: 1, 25-Dihydroxyvitamin D3 works as anti-inflammatory." *J Diabetes Res* 77 (2007): 47-57.
25. Hatanaka, Elaine, Patrícia Teófilo Monteagudo, Mauro Sérgio Martins Marrocos and Ana Campa. "Neutrophils and monocytes as potentially important sources of proinflammatory cytokines in diabetes." *Clin Exp Immunol* 146 (2006): 443-447.
26. Jagannathan, Bogdan, Madhumita, Marie E. McDonnell and Hyunjin Shin, et al. "Elevated proinflammatory cytokine production by a skewed T cell compartment requires monocytes and promotes inflammation in type 2 diabetes." *J Immunol* 186 (2011): 1162-1172.
27. Hotamisligil, Gökhan S., Narinder S. Shargill and Bruce M. Spiegelman. "Adipose expression of tumor necrosis factor-α: Direct role in obesity-linked insulin resistance." *Sci* 259 (1993): 87-91.
28. Hirosumi, Jiro, Gürol Tuncman, Lufen Chang and Cem Z. Görgün, et al. "A central role for JNK in obesity and insulin resistance." *Nature* 420 (2002): 333-336.
29. Minsheng, Yuan, N. Konstantopoulos and Lee Jongsoon. "Reversal of obesity-and diet-Induced insulin resistance with Salicylates or Targeted Disruption." *Science* 293 (2001): 1673-1677.
30. Hundal, Ripudaman S., Kitt F. Petersen, Adam B. Mayerson and Pritpal S. Randhawa, et al. "Mechanism by which high-dose aspirin improves glucose metabolism in type 2 diabetes." *J Clin Investig* 109 (2002): 1321-1326.

How to cite this article: Alfred Ehiaghe Friday, Udiyiwe Osaikhuwuomwan, Uwaifo Nicholas Eghosa and Ehiaghe Imuetinyan Joy, et al. "Antioxidant Trace Elements Depletion Induces Proinflammatory Cytokines Polarization in Anti Diabetics Drug Naive Non Obese Male Type 2 Diabetic Nigerians." *J Bioanal Biomed* 15 (2023): 364.