Comparative Study Of Some Immunological Aspects Between Type I And Type II Diabetic Mellitus In Iraqi Patients Of Thi-Qar Province.

Hayfaa A.Thijail BSc[1]. Hind M. Mousa PhD[2]

[1,2] *College of Since , University of Thi-Qar ,*

Email: Hindmousa_pa@sci.utq.edu.iq, Hindmousa155@yahoo.com

Abstract : This study designed to compare the vital role of the inflammatory immune responses and autoimmunity in development both types of diabetic mellitus by measurement of serum IL-17 , and Anti-Gad levels among patients groups and healthy by using ELISA kits . The study included sixty DM patients were divided into two groups : 30 patients with Type I DM (33.7%) , and 30 patients with Type II DM (33.7%) . In addition to 29 healthy individuals were (32.6 %) . The results indicated an elevated mean serum level of IL-17 and Anti-Gad in both patients groups , as compared with the healthy control with significant difference . Also, the findings recorded a highly significant difference in IL-17 and Anti-Gad sera levels between both types of patient groups at (P< 0. 01) . According to the correlation results between IL-17 and Anti-Gad in patients groups , the data was demonstrated that the expression of IL-17 had a significant positive correlation with expression of Anti-Gad (P< 0.05) among Type I patients . Whereas an inverse very weak correlation was observed between IL-17 and Anti-Gad with no significant (P > 0.05) in Type 2 patients . we concluded the an elevated levels of IL-17A in both types of DM may be as a prognostic factor for development them in Iraqi population. We also conclude that elevated Anti-Gad in Type II DM may be an indication of the rapid development of undiagnosed insulin deficiency in latent autoimmune diabetes in adult patients .

Key Words : Diabetes Mellitus ,IL-17, Anti-Gad-65 , Auto-immune diseases

1. INTRODUCTION

Diabetes mellitus (DM) is the major globally threatened metabolic disorder characterized by the presence of hyperglycemia, due to impaired or imbalanced insulin. It is a complex chronic disease that requires ongoing medical care with strategies to control blood sugar to reduce its multiple risks (IDF, 2017; ADA, 2020).

Type 1 diabetes is one of the most dangerous kinds of diabetes, as it affects about (5-10%) of all diabetes patients, and it occurs in early adolescence and in childhood and is also called (insulin dependent diabetes). It is recognized by a significant impairment of the functioning of pancreatic cells, which leads to an absolute decrease in insulin secretion, but type 2 diabetes is a chronic, multifactorial disease, reflects to a combination of environmental and genetic risk factors

It is accounted one of the most common kinds of diabetes, as it forms about (90-95%) of

all diabetes patients in the world. It happens that either the body does not use insulin as it should or does not produce enough insulin, leading to insulin resistance with a gradual loss of the ability to secrete beta cells to insulin (KC & Rao, 2013; ADA, 2020; Katsarou *et al.*, 2017; Zheng *et al.* ., 2018 ; Goyal & Jialal, 2019).

The cytokines have, a cardinal role in causing and exacerbating diabetes through direct and in direct mechanisms that cause to loss of the insulin producing beta cells of the islets of Langerhans in pancreas leading in to insulin deficiency (Ammon, 2019) . The levels of Some cytokines are associated with risk of developing diabetes mellitus such as interleukin(IL-17A) that produce by Th 17, which is abridge that links the adaptation and development the immune response and has important role in the (innate and adaptive immunity).

The recently identified interleukin 17 (IL-17) cytokines family play essential role in chronic inflammatory disease and host immunity against infectious disease (Liu, 2019) , and some studies have demonstrated association of (IL-17A) with pathogenesis of (Diabetes Type 1) (Honkanen *et al*., 2010; Linhartova *et al.*, 2016) and association with pathogenesis of diabetes Type 2 (Zareian and Dizgah , 2014). Pancreatic islet autoantibodies are proteins related to type 1 diabetes that released by the immune system (Burke *et al.*, 2020) . It is estimated that 10% in type 2 diabetes (T2D) patients have antibodies containing glutamic acid decarboxylase 65 (GADA) in the serum and these usually switch to insulin dependent patients within a few years, and are classified as latent Auto-immune diabetes in adults (LADA) (Åkesson *et al.*, 2010) .

2. MATERIALS & METHODS :

The current study was performed on ninety persons 60 of them that were patients suffering from diabetes mellitus, who attended the consultant clinic in endocrine and diabetic center in Al-Nassiriya city in the period from the beginning from September 2019 to March-2020. Blood samples were collected by venipuncture from 30 T1DM , 30 T2DM patients and 29 controls . Five ml of whole blood was collected in coagulate gel tubes and left to coagulate at room temperature, then for 10 minutes centrifuged at 3000 rpm to separate the serum . The serum was stored at -20 C˚ freezing. These sera (60 patients and 29 controls) were used for estimating the concentration of interleukin (IL-17 and Anti-GAD65) ELISA (technique enzyme-linked immune Sorbent adsorptive) kit are employing the quantitative sandwich ,were based on similar principle according to the (Bioassay Technology company ,china).

3. STATISTICAL ANALYSIS:

Data were expressed as mean ± standard deviation (SD). Differences between groups were tested with one way ANOVA test . The difference was considered significant when the probability (P) value was ≤ 0.05.Correlation coefficient was used as an indicator to express the relative relation between variables (Markers) and to measure the dependence of one variable on the other.

4. RESULTS & DISCUSSION :

4.1 : Serum Interleukin (IL17A) concentration of patients and Control :

The current findings demonstrated that there was a significant difference in sera mean levels of IL-17A among studied groups when they are analyzed statistically by ANOVA test($F = 6.935$, $P < 0.05$) .

The results indicated an increase in the serum level of (IL-17A) in patients with diabetes

of both types compared with the control group, as the concentration in serum of patients with diabetes type I (71.5985±16.575 Pg / ml),and its concentration in the serum of people with diabetes was the second type(64.1454±3.6288Pg / ml).Whereas in the control samples their concentration was (62,346±4.194Pg/ml). Also, the results appeared highly significant differences in IL-17A Sera level between both types of patient group at($P<0.01$), and between Type1DM and healthy ,($P<0.01$).While there was no significant difference between Type 2 DM patient and healthy control ($p=0.502$) as showed in the Table(1), Figure (1).

Table (1)The Levels of IL-17A in the study Groups.

Groups	No.	Mean± S.E. of IL-17 pg/ml	Comparison	Sig.
TYPE1 DM	30	71.5985±16.575	TYPE1 -TYPE2	0.005
TYPE2 DM	30	64.1454± 3.628	TYPE1-Healthy	0.006
Healthy	29	62.364 ± 4.194	TYPE2-Healthy	0.502

This result was relatively compatible with data reported by Nuhiar *et al* . (2018) in Thi-qar province. As well as the results were consistent with the results of both Roohi*et al*. (2014) and Reinert-Hartwall *et al*.(2015) who obtained high concentrations of IL-17 among the patients sample compared to the control sample.

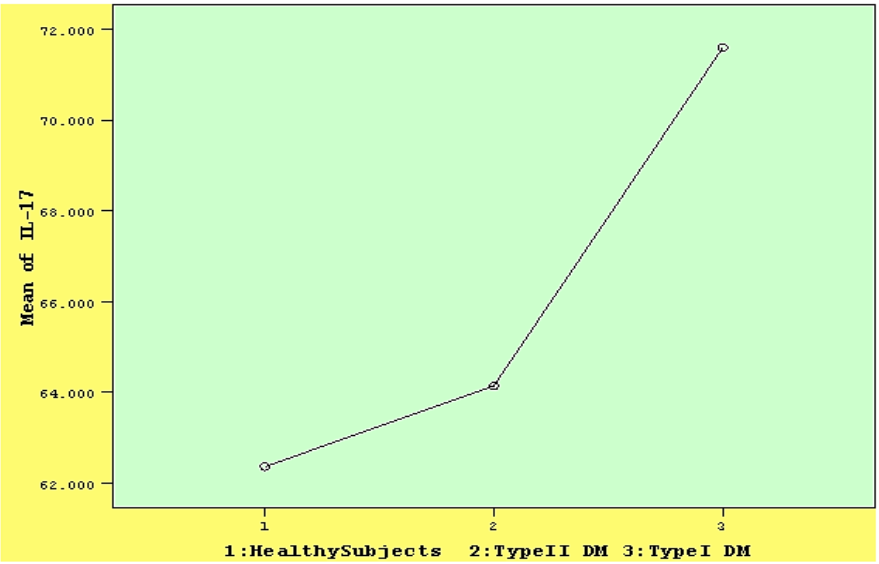


Figure (1) : The mean plot of IL-17A in study Groups.

Increasing evidence suggest that IL-17 plays a crucial role in various inflammatory responses and autoimmune diseases , most studies support the major role that (IL-17A) plays in the degradation of beta-producing insulin cells in the pancreas for people with diabetes – type1 . CD4 T lymphocytes differentiate into (Th17) cells in the tissue of the pancreas containing the cells that work to destroy beta cells by secreting them (IL-17A). Grieco *et al*. (2014) explained that IL-17A contributes to causing type 1 diabetes through two mechanisms, which included exacerbates the programmed death of the beta cell and increases production Chemokine's. (CXCL1, CXCL8) , that lead to exacerbate of inflammet and the death of pancreatic beta cells, so there is a link to a (IL-17A) and increased programmed death of beta cells.

This may mediate the harmful effect in the cells of the pancreatic islets in humans, the increase in the expression of cells (Th17) in people with type 1 diabetes generally acts as advanced indicators of the autoimmune response todegradation of beta cells, as the high production of IL-17A,It is observed when the autoimmune response against beta cells in patients with type 1 diabetes (He *et al*., 2014). Also, the finding of this study were consistent with a study conducted in Baghdad that included (50) samples of people with type 1 diabetes. This study found a significant increase in the level of (IL-17A) among patients compared to the control group (Tuama et al., 2014).While this result was not achieved with a study conducted to measure the level of (IL-17A) in patients with type I diabetes in the city of Baquba, which showed no significant differences between patients and the control group(Musleh and Ban, 2018) .

On the other hand , the results were in agreement with those of Borges et al. (2010) , who explained that the concentration of (IL-17A) was not different in patients with type 2 diabetes than the control group. The role of (IL-17A) in the pathogenesis and the events of the second type as cellular motor initiator of inflammation has been demonstrated, as the results of studies indicated that (IL-17A) is involved in the inflammatory process of type 2 diabetes and has a critical role in causing this pattern. Previous data found an elevated level of IL-17 in patients with type 2 diabetes, but the exact role of IL-17A in disease events was not discovered (Chen *et al*., 2016). Studies have confirmed that inflammation plays a critical role in insulin resistance and in the pathogenesis of type 2 diabetes (Carvalho *et al*., 2006)

IL-17 was considered to be toxic for cells , and it has a role in a cytostatic of islet cells in the pancreas that leads to inhibition of insulin production and secretion or stimulating production of nitrogen oxide . Also, this cytokine induces acute inflammation(Cieślak *et al.*, 2015).

4.2 : Serum(Anti-GAD65)concentration of patients of type 1and Type II Diabetes:

GADA is the most common antigen-specific autoantibody marker sensitive to autoimmune diabetes (Towns and Pietropaolo , 2011). The Present findings of ANOVA test pointed that there was significant difference among groups ($F = 86.538$, $P < 0.05$), where there was a significant increase ($p \leq 0.05$) in the levels of (anti-GAD)in both groups of DM patients (1. 8596, 1. 1952) as compare with control subjects (1.0588) Table (2) . This result was consistent with results reported by Basu *et al*. (2020) Who indicated a high significant increase in anti –GAD sera levels that it is found in about 79.3% of patients with Type I diabetes .Type I diabetes (T1D) is the result of loss of pancreatic beta cells that is driven by an underlying autoimmune disorder in which peripheral tolerance is lost , and mature auto reactive CD4 + and CD8 + cytotoxic T cells progressively destroy beta cells, supported by innate immune cells(Christoffersson , *et al.*, 2016).

Table (2) : Sera Levels of Anti-Gad in Patients and Healthy Groups.

DM Groups	**No.**	**Mean± Std. of Anti-Gad pg/ml**	**Comparison**	**Sig.**
TYPE1	30	1.8596±0.362	TYPE1 – TYPE2	0.00
TYPE2	30	1.1952±0.194	TYPE1-Healthy	0.00
Healthy	29	1.0588 ±0.135	TYPE2-Healthy	0.04

T1DM was previously the only type of immunodeficiency deliberated in Kids and adults alike, but in recent years, a number of researchers maintained, through their research, that there is another subtype called LADA with distinct researcher names as it has slow clinical characteristics when compared with T1DM and need insulin more quickly when compared with T2DM (Kumar and de Leiva, 2017). Anti-GAD also proved that it has a role in causing type 2 diabetes, where anti-GAD was shown to increase in T2DM Patients compare with controls, due to innate as well as adaptive immune (Hotamisligil, 2017; Němec and Zachariáš, 2018)

.

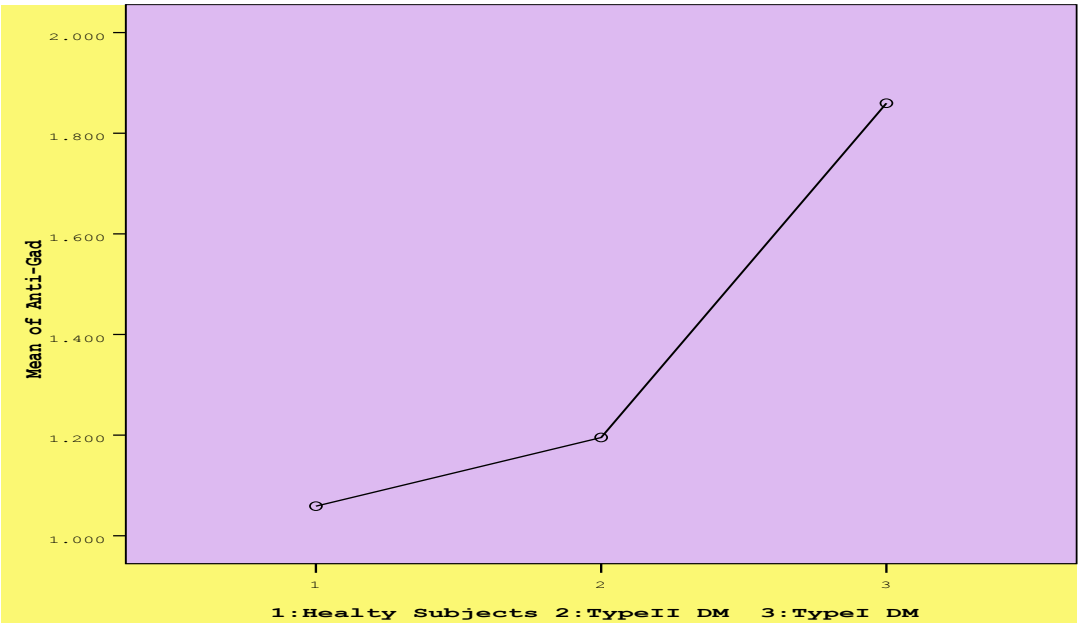


Figure (2): The mean plot of Anti-Gad in study Groups.

Autoimmunity of the islet cells in T2DM, chronic overfeeding and obesity lead to metabolic stress and adipocyte stress. Pro-inflammatory secretion by stressed adipocytes, such cytokines as IL-1β , IL-6 or TNF-α activate T- cells, B cells and macrophages. Overfeeding in particular, the increased concentration of circulating glucose and free fatty acids activate the inflammatory signaling which stimulates the production of IL-1β.Further macrophage recruite hyperglycemia that leads to a enhanced expression of several antigens of the β-cell, such as GAD, Consequently, increased β cell vulnerability to autoantibodies such as anti-GAD. Auto antibodies are the most commonly detected in phenotypic T2DM(Herawati *et al.*,2017).CD8 +T cells can be cytotoxic by attaching to MHC-I molecules on the pancreatic β- cell surface(Yeo *et al* .,2018).

T cells as well as macrophages all make a significant contribution to amplification of the inflammation of the islets by further discharge ofinflammatory Cytokines(Xia *et al.*,2017).

β cells up regulate expression of Fas in the presence of IL-1β and becoming more prone to apoptosis induced by activation of T cells via death domain signaling. Ultimately, cell apoptosis is the other result of all the inflammatory and autoimmune reactions, which cause a decrease of insulin concentration and worsening of hyperglycemia in patients. A common misdiagnosis for DM as a T2DM indicates that LADA patients are usually treated with medications mentioned for non-autoimmune forms of diabetes, leads to a rapid decrease in β-cell function, especially in younger , leaner subjects and those with incredibly high

GADA titers (Liu *et al*., 2019). The data by Lee *et al*. (2011) in Korean patients with type 2 diabetes reported that presence of GADA predicted progression to insulin deficiency.

4-3: Correlation coefficient between IL-17 & Anti-Gad parameters in patients

According to correlation coefficient between IL-17and Anti-Gad in patients groups , there was two different correlations between those biomarkers involved in patients Figures (3) and (4). Expression of IL-17 had a significant positive correlation with expression of Anti-Gad ($p<0.01$) among Type 1patients ,also an inverse very weak association was observed between IL-17 and Anti-Gad with no significant ($p \geq 0.05$) in type 2 patients . According to these results ,we found ,there was a positive correlation between IL-17 and Anti-Gad in type 1 diabetes mellitus. Grieco *et al*. (2014) clearly showed that IL-17A contributes to the initiation and progression of type 1 diabetes through two mechanisms, namely the exacerbation of beta cell apoptosis and increased local chemokine manufacturing, potentially worsening insulitis this finding agreed with fact suggested by Arif *et al*. (2011) who reported that peripheral blood CD4 T-cell from patients with type 1 diabetes mellitus release IL-17 in response to β-cell auto antigen.

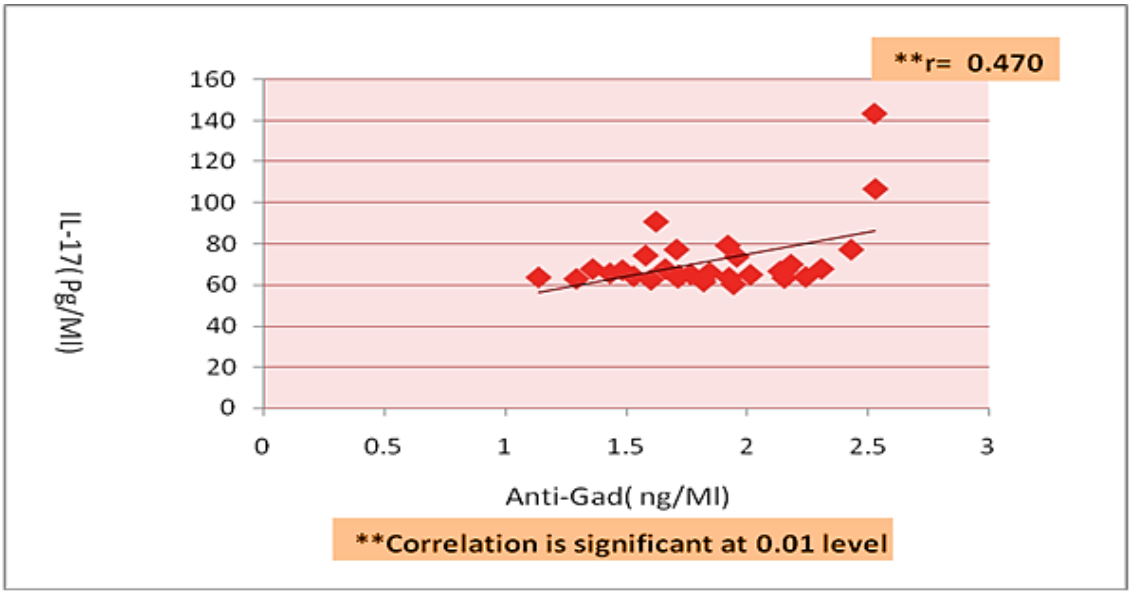
Also , this data pointed in human type I DM the direct role of IL-17 secreting cells in the etiological pathways to destroy β- cell . Direct viral, or a primary immune attack against specific β-cell antigens such as GAD65, may initiate damage to β-cells .T-helper lymphocytes (CD4 +) are activated by antigen-presenting cell and β-cell antigens. The activated T-helper cells produce cytokines that attract T and B lymphocytes and encourage them to proliferate in the islet leading to insulitis.Then B lymphocytes may damage β-cells by producing antibodies to released β-cell antigens(Hassan *et al*., 2012). Abd El-Latif *et al*.(2007) showed positive correlation between IL-17 and Anti-Gad.

On the other hand ,published study demonstrated that there was no correlation between the expression of IL-17A receptor and titles of pancreatic autoantibodies (anti-GAD65 or anti-IA2) (Fores , 2010).

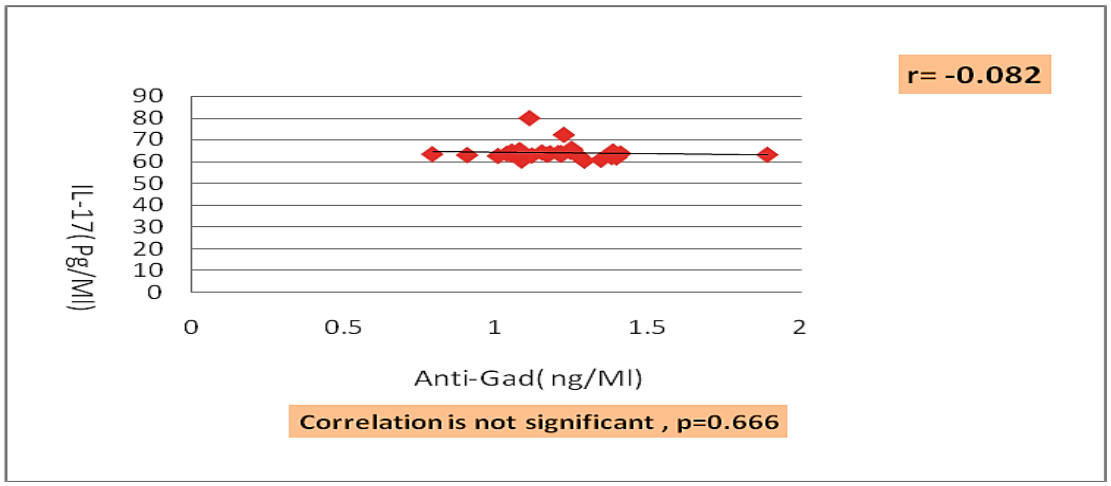
Table(3): Correlation between IL-17 &Anti-Gad in patients

IL-17 & Anti-Gad Correlation **Types of Disease**	**Pearson Correlation coefficient (r)**	**Level of sig.**
Type1 DM	**0.470**	**0.009****
Type2 DM	**- 0.082**	**0.666**

**Correlation is significant at the (P<0.01)level



Figure(3): The Positive Correlation between IL-17 & Anti-Gad in Type I DM Patients with significant difference .



Figure(4): The Negative Correlation between IL-17 & Anti-Gad in Type II DM Patients with no significant difference .

From all above finding about the concentration of inflammatory IL-17 and auto-antibodies , we concluded that the an increased concentration of IL-17A in both types of DM may be as a predisposing factor for promotion them in Iraqi population . We also conclude that elevated Anti-Gad in Type II DM may be a marker of the rapid development of undiagnosed insulin deficiency in latent autoimmune diabetes in adult patients .

5. REFERENCES

[1] **Abdel El-Latif, M., Abdel-Moneim, A. A., El-Hefnawy, M. H., & Khalil, R. G. (2017)**. Comparative and correlative assessments of cytokine, complement and antibody patterns in paediatric type 1 diabetes. *Clinical & Experimental Immunology*, 190(1), 110-121.

[2] American Diabetes Association ADA (2020) . Classification and Diagnosis of Diabetes: Standards of Medical Care in Diabetes-2020. *Diabetes Care*, 43(Suppl 1),S1.

[3] **Arif, S., Moore, F., Marks, K., Bouckenooghe, T., Dayan, C. M., Planas, R., ... & Huang, G. C. (2011)**. Peripheral and islet interleukin-17 pathway activation characterizes human autoimmune diabetes and promotes cytokine-mediated β-cell death. *Diabetes*, 60(8), 2112-2119.

[4] **Ates, I.(2018)**. Type 2 diabetes mechanisms, role of cytokines and their variations in disease development. *FABAD Journal of Pharmaceutical Sciences*, 43(1), 31-40.

[5] **Basu, M., Pandit, K., Banerjee, M., Mondal, S. A., Mukhopadhyay, P.,&Ghosh, S. (2020)**. Profile of auto-antibodies (Disease related andother) in children with type 1 diabetes. Indian Journal ofEndocrinology an Metabolism, 24(3), 256d. **.**

[6] **Borges, M. C.; Terra, G. A.; Takeuti, T. D.; Ribeir, B. M.; Silva, A. A., Terra - Junior, J. A.; Rodrigues -Junior, V. and Crema, E. (2010).** Immunological Evaluation Of Patients With Type 2 Diabetes Mellitus Submitted to Metabolic Surgery. *ABCD Arq Bras Cir Dig Original Article*, 28(4):266-269**.**

[7] **Braga, K.G.(2016) .**IL-6 and type 1 diabetes mellitus: T cell responses andincrease in IL-6receptor surface expression. Submitted Nov 17, 2016. Accepted for publication Nov 21.

[8] **Carvalho, C. and AL Fortes, Z.B. (2006*).Cy***tokines, Endothelial Dysfunction, and Insulin Resistance. *Journal Endocrinol Metabolism* , 50:304-12.

[9] **Chen, Z., Laurence, A., Kanno, Y., Pacher-Zavisin, M., Zhu, B. M., Tato, C., ... & O'Shea, J. J. (2006)**. Selective regulatory function of Socs3 in the formation of IL-17-secreting T cells. *Proceedings of the national academy of sciences*, 103(21), 8137-8142.

[10] **Christoffersson, G., Rodriguez-Calvo, T., & Herrath M. V. (2016).** Recent advances in understanding Type 1 Diabetes. *F1000Research*, 5: 1-9 .

[11] **Cielak, M. ;Wojtczak, A. and Cielak. M .(2015).** Role of pro-inflammatory cytokines of pancreatic islets and prospects of elaboration of new methods for the diabetes treatment. *Journal ActabiochimicaPolonica*, 62(1):15 -21.

[12] Diabetes Atlas, 8th Edition. https://doi.org/http://dx.doi. org/10.1016/S0140-

[13] diabetes patients are in Baqubah. Master Thesis, Faculty of Education forPure Sciences, De Yali University.

[14] **Doan, T., Melvold, R., Viselli, S., &Valtenbaugh, C. (2012).** *Immunology.* Lippincott Williams & Wilkins.

[15] **Domingo-Gonzalez, R., Prince, O., Cooper, A., &Khader, S. A. (2017).** Cytokines and chemokines in Mycobacterium tuberculosis infection. *Tuberculosis and the Tubercle Bacillus*, 33-72.

[16] **Fores, J. P. (2010).**Impacto da IL-17A napredisposiçãoao diabetes mellitus tipo 1A (Doctoral dissertation, Universidade de São Paulo).

[17] glucose tolerancein humans. *The Journal of Immunology*, 194(1), 68-75.

[18] Goyal, R.; Faizy,A.; Siddiqui,S. and Singhai,M. (2012). Evaluation Of Tnf-A And Il-6 Levels In Obese And Non obese Diabetics :Pre- And Postinsulin Effects .*North American Journal Of Medical Sciences*,4 (4),180-184**.**

[19] **Grewal, A. K., Arora, S., & Singh, T. G. (2019).** Role of Protein Kinase C in Diabetic Complications. *Journal of Pharmaceutical Technology, Research and Management*, 7(2), 87-95**.**

[20] **Grieco, F. A., Moore, F., Vigneron, F., Santin, I., Villate, O., Marselli, L., ... &Marchetti, P. (2014**). IL-17A increases the expression of

proinflammatorychemokines in human pancreatic islets. *Diabetologia*, 57(3), 502-511.

[21] **Grieco, F. A., Moore, F., Vigneron, F., Santin, I., Villate, O., Marselli, L., ... &Marchetti, P. (2014)**. IL-17A increases the expression of proinflammatorychemokines in human pancreatic islets. *Diabetologia*, 57(3), 502-511.

[22] **Grynberg, K., Ma, F. Y., &Nikolic-Paterson, D. J. (2017).** The JNK signaling pathway in renal fibrosis. *Frontiers in Physiology*, 8, 829**.**

[23] **Hanna, S. J., Powell, W. E., Long, A. E., Robinson, E. J., Davies, J., Megson, C., ... & Dayan, C. M. (2020).** Slow progressors to type 1 diabetes lose islet autoantibodies over time, have few islet antigen-specific CD8+ T cells and exhibit a distinct CD95 hi B cell phenotype. *Diabetologia*, 1-12.

[24] **Hassan, G. A., Sliem, H. A., Ellethy, A. T., & Salama, M. E. S. (2012).** Role

[25] **He, J. S., Xie, P. S., Luo, D. S., Sun, C. J., Zhang, Y. G., & Liu, F. X. (2014)**. Role of immune dysfunction in pathogenesis of type 1 diabetes mellitus in children. *Asian Pacific journal of tropical medicine*, 7(10), 823-826.

[26] **Herawati, E., Susanto, A., &Sihombing, C. N. (2017)**. Autoantibodies in Diabetes Mellitus. *Molecular and Cellular Biomedical Sciences*, 1(2), 58-64.

[27] **Hotamisligil, G. S. (2017)**. Inflammation, metaflammation and immunometabolic disorders. *Nature*, 542(7640), 177-185.

[28] *Indian journal of endocrinology and metabolism*, 16(6), 904.

[29] **International Diabetes Federation [IDF]. (2017).** Eighth edition 2017. IDF

[30] **Katsarou, A., Gudbjörnsdottir, S., Rawshani, A., Dabelea, D., Bonifacio, E., Anderson, B. J., ... &Lernmark, Å. (2017).** Type 1 diabetes mellitus. *Nature reviews Disease primers*, 3(1), 1-17.

[31] **KC, H. Y., & Rao, V. (2013).** Pharmacogenomics in diabetes mellitus: Pathway to personalized medicine. *Helix-The Scientific Explorer*, 2(6), 215-220.

[32] **Kumar, A., & de Leiva, A. (2017).** Latent autoimmune diabetes in adults (LADA) in Asian and European populations. *Diabetes/metabolism research and reviews*, 33(5), e2890

[33] **Lampasona, V., &Liberati, D. (2016).** Islet autoantibodies. *Current diabetes reports*, 16(6), 53.

[34] **Lee, K. U. (2011).** Progression to insulin deficiency in Korean patients with Type 2 diabetes mellitus positive for anti-GAD antibody. *Diabetic Medicine*, 28(3), 319-324.

[35] **Linhartova, P. B.; Kastovsky, J.; Lucanova,S.; Bartova, J.; Poskerova,H.;** Vokurka, J.; Fassmann,A.; Kankova, K.; and Holla, L. I. (2016). Interleukin-17A Gene Variability in Patients with Type 1 Diabetes Mellitus and Chronic Periodontitis: Its Correlation with IL-17 Levels and the Occurrence of Periodontopathic Bacteria. *Journal of Mediators of Inflammation*, 1-9

[36] Liu, W., Huang, X., Zhang, X., Cai, X., Han, X., Zhou, X., ... &Ji, L. (2019). Urinary C-Peptide Creatinine Ratio as a Non-Invasive Tool for Identifying Latent Autoimmune Diabetes in Adults (LADA). Diabetes, Metabolic Syndrome and Obesity: *Targets and Therapy*, 12, 2531.

[37] **Luopajärvi, K., Härkönen, T., ... &Tillmann, V. (2015)**. Th1/Th17 plasticityis a marker of advanced β cell autoimmunity and impaired

[38] **Ma, W. T., Chang, C., Gershwin, M. E., &Lian, Z. X. (2017).** Development of autoantibodies precedes clinical manifestations of autoimmune diseases: a comprehensive review. *Journal of autoimmunity*, 83, 95-112.

[39] **Meffre, E., & O'Connor, K. C. (2019)**. Impaired B-cell tolerancecheckpoints promote the development of autoimmune diseasesandpathogenic autoantibodies. *Immunological Reviews*, 292(1), 90–101.

[40] **Musleh. Ban S . (2015).** Some immunological indicators in type I diabetes in Baqubah

. Master Thesis, College of Education for pure Sciences, University of Diyala .

[41] Němec, M., &Zachariáš, J. (2018). The KrásnáHora, Milešov, and PříčovySb-Au ore deposits, Bohemian Massif: mineralogy, fluid inclusions, and stable isotope constraints on the deposit formation. MineraliumDeposita, 53(2), 225-244

[42] **Nuhiar R. S. , Salman A.N. , and AL-Rekaby H. R. (2018).** The some cytokines Levels (TGFβ1, IL-4 , IL-6, and IL-17) in sera Patients with Diabetes Mellitus Type1,Type 2 in Nassiriya city . *J.Thi-Qar Sci.*, 6(4):42-48.

[43] **Reinert-Hartwall, L., Honkanen, J., Salo, H. M., Nieminen, J. KRojas-Carranza, C. A., Bustos-Cruz, R. H., Pino-Pinzón, C. J., Ariza-Marquez, Y. V., Gómez-Bello, R. M., &Canadas-Garre, M. (2018).** Diabetes-related neurological implications and pharmacogenomics. Current pharmaceutical design, 24(15), 1695-1710.

[44] **Theofilopoulos, A. N., Kono, D. H., &Baccala, R. (2017).** The multiple pathways to autoimmunity. *Nature immunology*, 18(7), 716**.**

[45] **Towns R. and Pietropaolo M. (2011).** GAD65 autoantibodies and its role as biomarker of Type 1 diabetes and Latent Autoimmune Diabetes in Adults (LADA). *Drugs Future. Nov*; 36(11): 847.

[46] **Tuama , A.; Hassan, H. and Rashid ,K. (2014).** Estimation the levelof il-17a in a sample oftype i diabetes mellitus patients. *IOSR Journal Of Pharmacy (p)-ISSN*: 2319-4219: 57-60.

[47] **Xia, C., Rao, X., &Zhong, J. (2017).** Role of T lymphocytes in type 2 diabetes and diabetes-associated inflammation. *Journal of diabetes research*, 2017.**.**

[48] **Yeo, L., Woodwyk, A., Sood, S., Lorenc, A., Eichmann, M., Pujol-Autonell, I., ... &Tungatt, K. (2018)**. Autoreactive T effector memory differentiation mirrors β cell function in type 1 diabetes. *The Journal of clinical investigation*, 128(8), 3460-3474.

[49] **Zareian, P., & Dizgah I. M. (2014).** Serum interleukin 17 in type 2 diabetes mellitus. *Journal of Archives in Military Medicine*, 2(4).e24986.

[50] **Zheng, Y., Ley, S. H., & Hu, F. B. (2018).** Global etiology and epidemiology of type 2 diabetes mellitus and its complications. *Nature ReviewsEndocrinology*, 14(2), 88.