Research Article

Selenium Synergize Levothyroxine in Restoring Leukocytes Cluster Differentiation Expression in Methimazole Induced Hypothyroidism

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Abstract | Thyroid hormones and selenium has been shown to play essential role in immune system, however the impact of hypothyroidism in immune cells and its relationship with selenium supplement in vivo has not well explored. Accordingly the present study designed to elucidate role of selenium in immunosuppression of hypothyroidism rats. Forty four female Wister rats of 3-4 months aged were equally and randomly divided into 5 groups except the control, which contains 8 rats. The 2nd, 3rd, 4th, and 5th groups were subjected to hypothyroidism by oral intubation of 0.02% Methimazole daily for 4 weeks. Treatments of 3rd with Sc-SeNPs (0.1mg selenium /kg/ day) as T1, 4th with Levothyroxine (0.9 µg/100g BW/day) as T2, 5th with SC-SeNPs (0.1mg selenium /kg/ day)+ Levothyroxine (0.9 µg/100g BW/day) as T3, in addition to 2nd hypothyroidism untreated group for 4weeks. Immunohistochemistry analysis showed that expression of CD4, CD8, and CD19 on Tand B lymphocytes and CD68 on macrophages were affected inversely by hypothyroidism. Selenium supplementation synergies levothyroxine in reestablishment the activity of lymphocytes and macrophages. In conclusion, the present study demonstrated that thyroid hormones had regulatory role for innate and adaptive, and in order to reach the best result of treating hypothyroidism and improving the activity of the immune cells , selenium must be given with the Levothyroxine

Keywords | Primary Hypothyroidism, SC-SeNPs, Levothyroxine, lymphocyte, CD, Nano-biotechnology , Antioxidants ,and IHC

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INTRODUCTION

Hypothyroidism from endocrinologically point is low plasma levels of T4 and T3 and high plasma levels of thyroid stimulating hormone (TSH) which is a condition that is quite common around the world. Hypothyroidism is described in multiple animal species, including cat, dog, pig, horse, sheep, goat and chicken (Annemarie et al., 2016; Gurda et al., 2017; Rustam et al., 2021; Stenlund, 2022).The hypothyroidism either thyroid dysgenesis or dyshormogenesis (Broome et al., 2019; Van Poucke et al., 2022). Reduction in circulating thyroid hormones slows cell metabolism and growth (De Leo et al., 2016); memory impairment, and neurological disorder in dogs (de Oliveira et al., 2022). Hypothyrodism reduced the health related quality of life in addaition to depress and mood disturbance (Ellegård et al., 2021; Ramezani et al., 2023). In comparison to men, women are five to eight times more likely to develop hypothyroidism (Meng et al., 2015). This prompted researchers to study the details of the effect of the condition on offspring. Recently maternal hypothyrodism caused neurological and brain function alteration of

rats offspring spring (Madhusudhan et al., 2022; Yan-Jun et al., 2023; Sabah Farhat et al., 2023).

In hypothyroidism, both innate and adaptive immune system were affected. Immune system activity is modulated by thyroid hormones, particularly cellular immunity. Hypothyroidism's immune system dysfunction may make infections more likely. Except for a reduction in the amount of natural killers in lower thyroid volumes and the high levels of antithyroid antibodies suggesting the activity of the autoimmune process, thyroid volumes in individuals with autoimmune thyroiditis are not related with substantial changes in cellular immunity (Al-Mendalawi, 2018; Goncharova, 2020) But with appropriate hormone replacement therapy, the immune system returns to normal (Mitrou et al., 2011). In severe hypothyroidism, T lymphocyte subtypes are activated, natural killer cells are decreased, and T CD4 lymphocyte responsiveness is decreased (Volpé, 2001).

For people with hypothyroidism, levothyroxine replacement therapy is the standard of care. However, a sizable portion of levothyroxine-treated patients continue to experience symptoms despite achieving biochemical therapy targets, raising the question of whether levothyroxine treatment is adequate for all patients or whether alternative therapies are necessary (El Deib et al., 2021). In severe hypothyroidism, T lymphocyte subtypes are activated, natural killer cells are decreased, and T CD4 lymphocyte responsiveness is decreased (Volpé, 2001; Ahmed et al., 2012).

Selenium (Se) is a trace element that is needed for the healthy operation of the thyroid and immune systems. The principal way that selenium affects thyroid control is through selenoproteins, a broad family of enzymes that have a significant impact on the oxidative balance, cell division and proliferation, and the generation of thyroid hormones (Duntas, 2006). While studies have shown that increased selenium can have some effects on immunological processes, (Mohan and Damodar, 2019) there is minimal evidence that it can also affect lymphocyte proliferation and activity in hypothyroidism conditions 1.The aims of the study to elucidate the defects in immune system activities and component. In hypothyroidism experimentally induced in female rats.

MATERIAL AND METHODS

Preparation of Se-nanoparticles

The biogenic synthesis of Se-NPS was achieved using saccharomyces cerevisiae as reducing agent following procedure of (Hariharan et al, 2012; Wali, 2019; Al-Kurdy, 2020) with some modifecations. Briefly: 100 gm of dried yeast dissolved in 1 L of deionized distilled water at 45C° in conical flask, and stirring with 50 gm sugar by 500 rpm stirring power for one hour, then, the yeast solution was mixed with 0.1 M of sodium hydrogen selenite (NaHSeO3) solution in 2:1 ratio v/v. Finally, the flask was stirred again on the magnetic stirrer (200rpm) at room temperature (22-24 C°) a for 24 hours and monitoring the formation of red brick colour. which is the initial sign of the generation of selenium nanoparticle (SeNPs). The prepeared selenium nanoparticles (Sc-SeNPs) solution was sonicated for 10 mins and washed twice with DDW after that dried the Sc-SeNPs solution in rotary evaporator. The dose of Sc-SeNPs was fixed according to the concentration of Se elements in the preapared nanoparticles by atomic absorption and 0.1 mg of Se /kg body weight (Wali and Alqayim 2019).

Experimental Design

Forty four female Wister rats of age ranged between three to four months were randomly divided into two groups. The first group (8 rats) had daily been fed basal diet and given normal water for one month (control group), the second, third, fourth and fifth groups (36 rats) had daily been fed basal diet and given 0.02% Methimazole in drinking water for 4 weeks to induce hypothyroidism (Sabea and Alqayim 2023) . The hypothyroidism experimental rats of 2nd group untreated, 3rd, 4th, 5th, groups treated with Sc-SeNPs (0.1mg selenium /kg/ day)as T1, or Levothyroxine (0.9 µg/100g BW/day) as T2, or SC-SeNPs (0.1mg selenium /kg/ day)+ Levothyroxine (0.9 µg/100g BW/day) as T3 for 4 weeks. After end the experiment, the animals were anaesthetized using a mixture solution composed of Ketamine and Xylazine with 90 and 10 mg/kg/B.W, respectively, intra-peritoneal (IP) (Lei et al., 2001). Blood sample were collected from anesthetized rats via cardiac puncture. Mesenteric lymph node and Spleen , isolated and immediately preserved in neutral buffer then in 10% formalin solution.

Cluster Differentiation

Within 24 hours Paraffin embedded lymphoid tissue sample were sectioned at 4 µm thickness and placed carefully in water bath and mounted on positive charged glass slides using a hot plate. The tissue sections were rinsed in distilled water and immersed in Tris-buffered saline (TBS) bath for 5 minutes. Then section put in glass jar filled with antigen retrieval solution pre-heated at 60 oC and incubated in a water bath at 97 oC for 25 minutes, the Kit that used Dako EnVision detection code K8000 .The primary applied antibodies were CD4, CD8 and CD19, and CD68 in lymphoid tissue as well as the antibody dilutent factor (code number K800). The antibodies utilized in the current investigation are displayed in Table 2.

The secondary antibody goat, anti HRP conjucated rabbit anti mouse.

Table 1: Primary antibody used in the study

Primary antibody	Host	Dilution factor	Marker	Cat. No/	Brand, Country
CD4	Rabbit, polyclonal	200x	T.helper lymphocyte	E-AB-22098	Elabscience, USA
CD8	Rabbit, polyclonal	100x	T.cytotoxic lymphocyte	E-AB-65844	Elabscience, USA
CD19	Rabbit, polyclonal	100x	B cell	E-AB-60746	Elabscience, USA
CD68	Rabbit, polyclonal	100x	Macrophage	E-AB-64533	Elabscience, USA

Table 2: Thyroid hormones in hypothyroidism female rats treated with Sc-SeNPS alone or in combination with thyroxin rats for 4weeks

GROUP	T3(nm/l)	T4(nm/l)	TSH(ng/ml)
Control	1.2±0.08A	35.49±2.89A	2.38±0.29C
Hypothyroidism	0.46±0.05B	7.06±0.77C	5.58±0.2A
T1	0.46±0.05B	14.37±2.76B	4.81±0.17B
T2	1.22±0.16A	36.39±2.23A	2.52±0.29C
T3	1.02±0.09A	30.26±2.57A	2.75±0.41C
LSD	0.27	6.99	0.84

Different capital letters denote significant differences between groups. T1:hypothyroidism and treated with Sc-SeNPs 0.1 mg / kg B.W, T2 hypothyroidism and treated with Thyroxin 0.9 μg/100g BW.day, T3: Hypothyroidism and treated with thyroxin+ Sc-SeNPs 0.9 μg /100g BW. day, and 0.1 mg /kg B.W.

RESULT AND DISCUSSION

Assessment Of Hypothyroidism

The current findings, which are shown in Table 1, showed that treatment of hypothyroidism in rats with a combination of levothyroxin and SC-SeNPs produced the best thyroid hormone levels when compared to hypothyroidism treated with SC-SeNPs alone and untreated. T4 was found to be the biomarker most adversely influenced by the Integrated Biomarkers response (Figure 1) analysis.

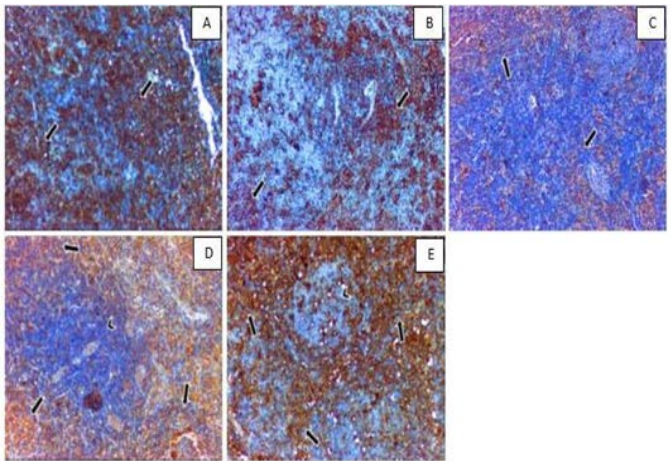

Figure 1: Photomicrograph of mesenteric (paracortex area) lymph node. Positive expression of T-lymphocyte helper receptor (CD4) (arrow) is shown in (A) control group, (B) hypothyroidism group, (C) SC-SeNPs, (D) levothyroxine group, and (E) SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 100x. Note that the lymphoid follicle (arrowhead) in (D and E) did not show CD4 expression.

The definition of hypothyroidism is mostly biochemical because to the wide variance in clinical presentation and general lack of symptom specificity. TSH levels that are above the guideline range and free thyroxin levels that are below the reference range are referred to as overt or clinical primary hypothyroidism. TSH concentrations above the reference range and free thyroxin concentrations within the normal range are the criteria for mild or subclinical hypothyroidism, which is sometimes viewed as a symptom of early thyroid failure. Normal thyroid hormone levels along with higher amounts of thyroid stimulating hormone are indicative of subclinical hypothyroidism..Thyroid hormones, L-thyroxine (T4) and 3,3′,5-triiodo-L-thyronine (T3), appear to be immune response modulators, according to growing research. Under hypo- and hyperthyroid circumstances, immunological functions such as chemotaxis, phagocytosis, reactive oxygen species (ROS) formation, and cytokine synthesis and release are altered in monocytes, macrophages, leukocytes, natural killer cells, and lymphocyte (De Vito, 2012; Ghazi and Al-Qaiym, 2023). Immunosuppression is well known to be caused by hypothyroidism (Al-zyadi,2015; Paulazo, 2019; Fakri et al 2023).

The result of Figures (1-2),(3-4) showed Mesenteric lymph node for CD4 and CD8 in hypothyroidism and SC-SeNPs groups rats.Positive expression of T-lymphocyte helper receptor (CD4) that covered less than 50% of Paracortex area of lymph node. Figures (5-6) showed CD19 expression in mesenteric lymph node, Weak and moderate in hypothyroidism and SC-SeNPs group rats, expression of B-lymphocyte antigen (CD19) in cortex area, where the

expression was observed in follicle center. Figures (7-8) showed weak and moderate expression of anti-CD68 primary antibody for hypothyroidism and SC-SeNPs group rats in red pulp area indicating the low phagocytic activity of macrophage compared with expression of anti-CD68 primary antibody in control group. Figures (9) showed the scoring for CD4, CD8, CD19 and CD68

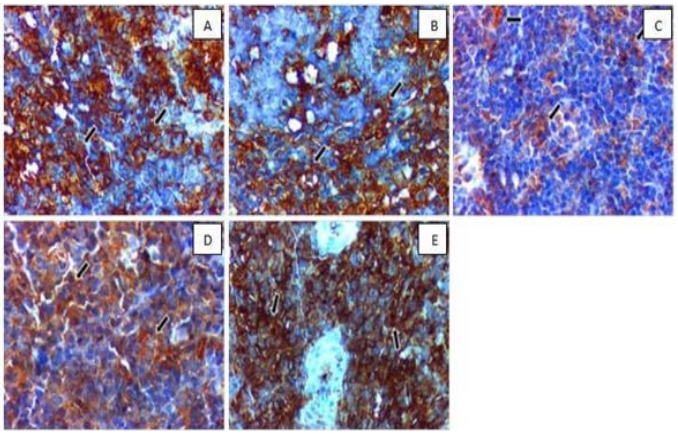

Figure 2: Photomicrograph of mesenteric (paracortex area) lymph node. Positive expression of T-lymphocyte helper receptor (CD4) (arrow) is shown in (A) control group, (B) hypothyroidism group, (C) SC-SeNPs, (D) levothyroxine group, and (E) SC-SeNPs+levothyroxine group.

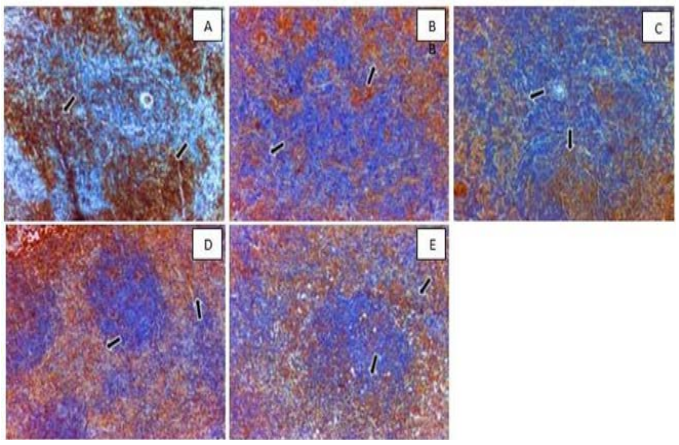

Figure 3: Photomicrograph of mesenteric (paracortex area) lymph node. Positive expression of T-lymphocyte helper co-receptor (CD8) (arrow) is shown in (A) control group, (B) hypothyroidism group, (C) SC-SeNPs, (D) levothyroxine group, and (E) SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 100x.

The present study aimed to shed light on the effects of hypothyroidism on the immune activity of some white blood cells through the detection of the antigen present on their surface by immunohistochemistry. The relationship between thyroid gland and immune system can be likened to to-sided coin. One side that thyroid hormone and thyroid stimulating hormone receptors are located on the surface of the B and T lymphocytes, monocytes and dendritic cells the Antigen presenting cells (Berczi et al., 2005; Adamczewski et al., 2020).

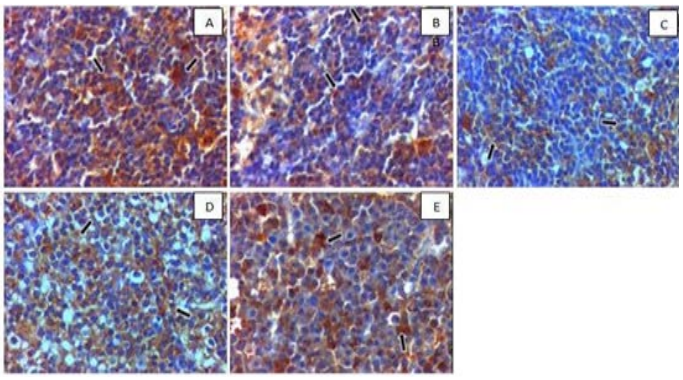

Figure 4: Photomicrograph of mesenteric (paracortex area) lymph node. Positive expression of T-lymphocyte helper co-receptor (CD8) (arrow) is shown in (A) control grou , (B) hypothyroidism group, (C) SC-SeNPs, (D) levothyroxine group, and (E) SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 400x.

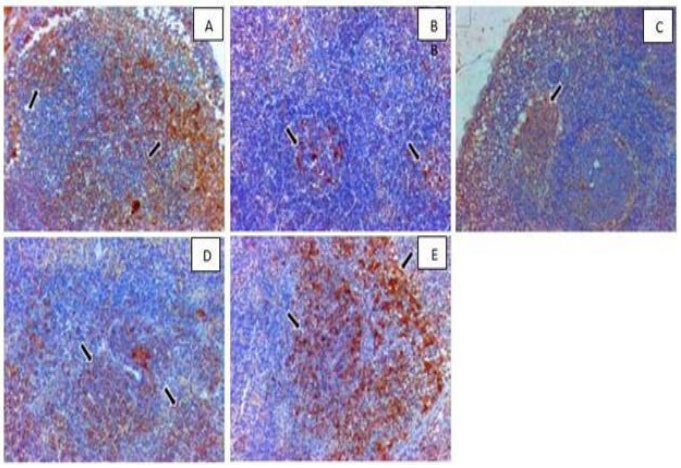

Figure 5: Photomicrograph of mesenteric (cortex area) lymph node. The expression of B-lymphocyte antigen (CD19) (black arrow) has shown (A) positive expression in control group, (B) weak expression in follicle area of hypothyrodism group, (C) moderate expression in the SC-SeNPs group, (D) positive expression in the levothyroxine group, and (E) positive expression in the SC-SeNPs+levothyroxine group.

The other side deal with effect of immunity on thyroid disease as autoimmune disease (Mansourian et al., 2010; Felicetti et al., 2017), stimulation of immunity by probiotics enhance thyroid (Alqayim,2015). The cluster differentiation (CD) are antigens located on the surface of cells, in physiological term it may excert activity as receptors, lignds that acti vate receptors(coreceptor) mediate cell signals and other functions (Guidebook. 2004). Cells that express CD4 are T-helper and regulator cells, mediate cellular immunity and delayed inflammation through their capability to produce humoral inflammatory cytokines.

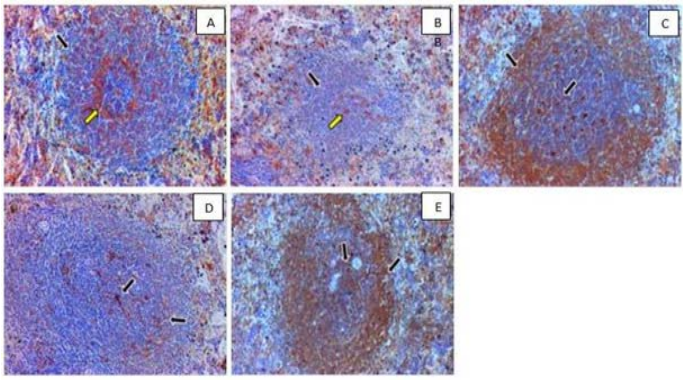

Figure 6: Photomicrograph of lymphatic follicle of white pulp in spleen. The expression of B-lymphocyte antigen (CD19) (black arrow) has shown (A) positive expression with intensive quantity (yellow arrow) at the center in control group, (B) weak expression in follicle area with intensive quantity (yellow arrow) at the center of hypothyrodism group, (C) positive expression in the SC-SeNPs group, (D) low-intensity expression in the levothyroxine group, and (E) positive expression in the SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 100x.

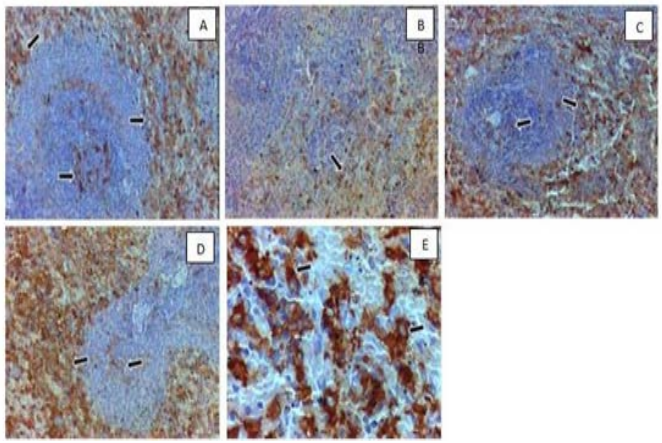

Figure 7: Photomicrograph of lymphatic follicle of red pulp in spleen. The expression of anti-CD68 primary antibody (arrow) has shown (A) positive expression in control group, (B) weak expression in hypothyrodism group, (C) moderate expression in the SC-SeNPs group, (D) strong expression in the levothyroxine group, and (E) strong expression in the SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 100x.

In the present study CD4, found to decreased significantly with increase of TSH, in on other word CD4 are decreased with thyroid dysfunction (Mathur et al., 2020). The relationship between thyroid dysfunction as reduced T3 and T4 and CD4 was well studied in pathological conditions, such as HIV infected persons (Indikar & Shoukat, 2019). In spite of the big volume of researchs that correlate the hypothyroidism with reduced CD4 (Dev et al., 2015; Janyga et al., 2021), but little are concerned with cellular mechanism for this reduction. the present study results of CD4 immunohistochemistry detection in lymph node revealed that decrease in CD4 resulted from a decrease in thyroid hormones. Because The expression of CD4 on variable cells this means that it's decrease with several effects, and it may not involve in the pathogenesis of AIT (Mekova et al., 2020) but on the contrary it resulted from T3 and

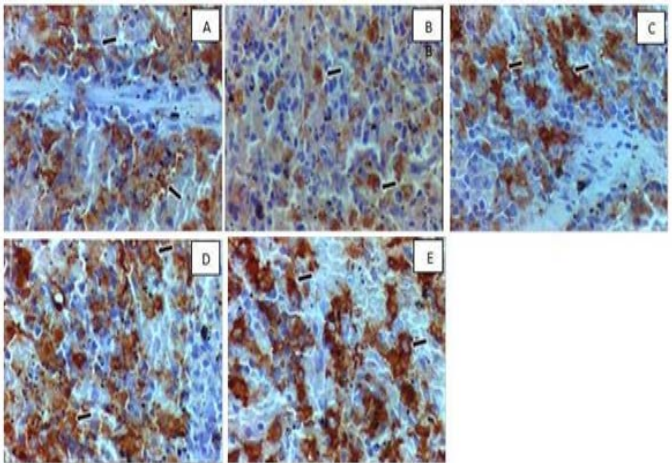

Figure 8: Photomicrograph of lymphatic follicle of red pulp in spleen. The expression of anti-CD68 primary antibody (arrow) has shown (A) positive expression in control group, (B) weak expression in hypothyrodism group, (C) moderate expression in the SC-SeNPs group, (D) strong expression in the levothyroxine group, and (E) strong expression in the SC-SeNPs+levothyroxine group. Hematoxylin & DAB. 400x

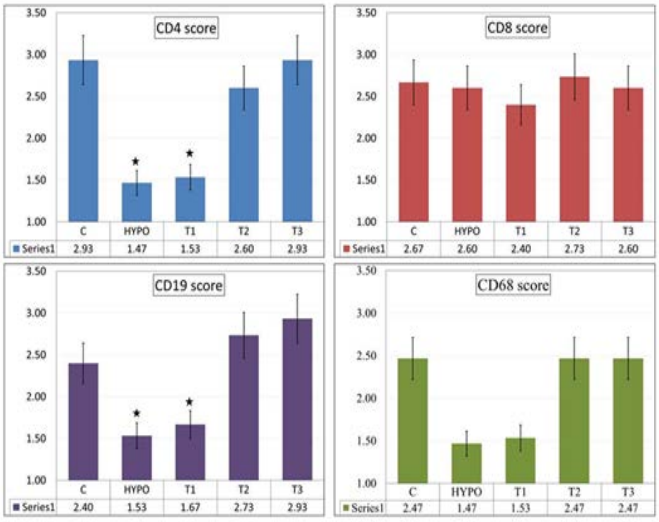

Figure 9: Scoring for CD4, CD8, CD19 and CD68

T4 deficiency. However this subject is contraversal because recently found that genetically TRH overexpression mice showed no different picture for CD4 (Valli et al., 2023), however . Measurment of CD4 immune cells is of diagnostic value to evalute the immune deficiency (Shete et al., 2010). The present reduction in CD4 expression moniter the immune deficiency in methimazole induced hypothy-

roidism. The differentiation of lymphocytes to be CD4 types is mediated by antigen presenting cells expressed major histocomtability class II, Activated rat T cells have been reported to express MHC class II molecules (Benoist et al., 1990; Mangalam et al., 2006), the instance of CD4 differntiation from Tcells is highly dependent of affinity of Tcell to MHC class II (Lagattuta et al., 2022). This un well defined CD4 differentiation in lymphoid tissue of the present hypothyrodism rats could be attributed to the alteration in the MHC-II caused by reduced thyroid hormones. Recently founded that aging thyrocytes that showing functional deterioration associated with the expression of metallothionein (MT) and major histocompatibility complex class II genes (Hong et al., 2023), may be due to decrease in thyrotropic cells in production for thyroid hormones (Lewiński et al., 2006; Gauthier et al., 2020).The CD8+ T cells are cytotoxic T cells that are primarily responsible for immune defense against intracellular pathogens and tumor monitoring, and participate in aoutoimmune disease (Abbas et al., 1996; Suzuki, et al., 2008). The non-significant reduction in CD8 expresion in lymphoid tissue of hypothyrodism rats could be attributed to the low sensitivity of Tcells to the antigen presented by antigen presenting cells with MHC class I (Shihab et al., 2021; Daniels et al., 2001). The CD8 cytotoxic cells are important effector in cell mediated immunity specifically against intracellular pathogen like viruses.The immunohistochemistry evaluation of CD19 in lymphoid tissue was performed to explore the effects of hypothyroidism model by trimethmazole as non-autoimmune. The CD19 one of the B cells is an important part of the adaptive immune system, largely because of their ability to mature into plasma cells and produce antibodies. They also present antigens and secrete cytokines (Maecker et al., 2012). The expression of these plasma subset cells are higher with higher thyroid hormones (Liu etal., 2018). The CD19 overexpressed by high level of thyroid hormones (Wang et al., 2021; Cao et al., 2022). The down regulation of CD19 expression in the present model of hypothyroidism indicated the non immunopathy condations in our experiment, because CD19 are up regulated in tumor disease (Poe et al., 2012). Whereas the down regulation of these cells is target in treatment of immunopathy disease, represented by disturbance in cell proliferation and apoptosis (Wu et al., 2018). Thyroid hormones approved for their immunomodulation activity, this modulation via over the and the over expression of B- cell activating factor with high thyroid hormones . The detection of CD68 expresion cells in lymph node was performed in attempt to evaluate macrophage phagocytosis activity. Triggering receptor expressed on myeloid cells-2 (TREM2) and CD68 cell surface protein are expressed is the mechanism by which start cellular signal pathways for anti-inflammatory phagocytic activity. Thyroid hormone positively regulate the expression of the TREM2 (Ferrara et al., 2022).The present results of immunohistochemistry detection of immune cells depending on their cell surface molecules expression clearly demonstrated the influence of thyroid hormone deficiency on immunity. In the present model of methimazole induced hypothyroidism were no pathogenic infection, the Tand B cell proliferation and differentiation are expected, but we don't agnore the possible role of thyroid hormones deficiency in the immune-related adverse events (Montesinos et al., 2009; Shintaro et al., 2022). However the limitation of the present works that this immune deficiency in hypothyroidism rats has not been studied in pathological and infectious condition. The addition of Se- nanoparticles with no thyroid hormones replacement did not show any effects on the CD4 expression. This is self-evident, as selenium does not compensate for the deficiency of thyroid hormones, but rather it is limited to antioxidant activity in the present study. On the contrary in groups with levothyroxine treatment, replacement of thyroid hormones worked to compensate for the deficiency of these hormones and thus reverse the possible changes in MHC-II and the return of CD4 differentiation to normal conditions. Best solution for this immune disorder in hypothyroidism is to compensate for the thyroid hormone deficiency with replacement. An increase to confirm this fact was the immunohistochemistry results of CD8 expresion in groups treated with levothyroxine and Sc-SeNPs.

CONCLUSION AND RECOMMENDATIONS

The present study demonstrated that thyroid hormones had regulatory role for innate and adaptive, and in order to reach the best result of treating hypothyroidism and improving the activity of the immune cells, selenium must be given with the Levothyroxine.

It's crucial to conduct more research to determine how can immune system response to infections in hypothyroidism condition, and here the role of selenium can be clear.

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CONFLICT OF INTEREST

There is no conflict of interest.

NOVELTY STATEMENT

This work has highlighted the role of selenium synergize levothyroxine in restoring leukocytes cluster differentiation expression in Methimazole induced hypothyroidism in rats.

AUTHORS CNTRIBUTION

Majida A. J. Al-Qayim designed the study and analyzed data. Aryaf Mahmood Sabea did the experiments and wrote the paper.

REFERENCES

Aati E, Al-Ali Z (2020). Effect of hypothyroidism on lipid profile in women at Misan City/Iraq. Medical J. Babylon., 17: 1-5. https://doi.org/10.4103/MJBL.MJBL_86_19

Abbas AK, Murphy KM, Sher A (1996). Functional diversity of helper T lymphocytes. Nature, 383: 787-793. https://doi.org/10.1038/383787a0

Adamczewski Z, Stasiołek M, Zygmunt A, Śliwka PW, Wieczorek-Szukała K, Lewiński A (2020). Recombinant Human Thyroid-Stimulating Hormone Increases the Percentages of Natural Killer T Cells and B Lymphocytes in Human Peripheral Blood In Vivo. Front Endocrinol (Lausanne), 11:543845. https://doi.org/10.3389/fendo.2020.543845

Ahmed N, Khalil E, Ghazi H, AL-Shami A, Hamza S, Al-Faisal A (2012). Detection of CD3, 19 and CD4, 8 markers on lymphocyte of lymphocytic leukemia Iraqi patients. Sec. Scient. Confer.,: 111-115.

Al-Kurdy M, Khudai KK, Al-Kinani LH (2020). Synthesis and characterization of black currant selenium nanoparticles (Part I). Iraqi J. Vet. Med., 44(2): 25-34. https://doi.org/10.30539/ijvm.v44i2.974

Al-Mendalawi MD (2018). Thyroid abscess: A rare case report and review of literature. Thyroid Res. Pract., 15(2):99.

Alqayim M (2015). Effects of Lactobacillus acidophilus on Pituitary-thyroid axis in growing rat. Adv. Anim. Vet. Sci., 3(5): 269-275. http://dx.doi.org/10.14737/journal.aavs/2015/3.5.269.275

Al-Shaikh M, Wahab T, Kareem S (2014). Hypothyroidism induced by carbimazole in diabetic mice and Its management using parsley and Eruca sativa oil. IOSR J. Pharm. Biolog., 9(1): 24-27. https://doi.org/:10.9790/3008-09152427

Al-Zyadi AJH, Arrak JK (2015). The therapeutic role of alcoholic extract of fenugreek seeds on hypothyroidismstate induced by thiourea and some blood parameters in adult male rabbits. Iraqi J. Vet. Med., 39(1): 1-7. https://doi.org/10.30539/iraqijvm.v39i1.187

Annemarie M, Peter A, Jenny J, Sylvie D, Hans S (2016). Central Hypothyroidism in Miniature Schnauzers. J. Vet. Inter. Med., 30(1): 85-91. https://doi.org/10.1111/jvim.13818

Arcos M, Cremaschi G (2019). Hypothyroidism-related zinc deficiency leads to suppression of T lymphocyte activity. Endocrine., 66: 266-277. https://doi.org/10.1007/s12020-019-01936-7

Bao-Kang Wu, Qian-Hui Chen, Dan Pan, Bing Chang, Li-Xuan Sang (2021). A novel therapeutic strategy for hepatocellular carcinoma: Immunomodulatory mechanisms of selenium and/or selenoproteins on a shift towards anti-cancer. Int. Immunopharmacol., 96:1567-5769. https://doi.org/10.1016/j.intimp.2021.107790

Benoist C, Mathis D (1990). Regulation of major histocompatibility complex class-II genes: X, Y and other letters of the alphabet. Ann. Rev. Immunol., 8(1): 681-715. https://doi.org/10.1146/annurev.iy.08.040190.003341.

Berczi I (2005). Neuroendocrine Regulation of Natural Immunity. In: L. Bertók and D. A. Chow (Eds.), NeuroImmune Biology, Elsevier, 5:215-262. https://doi.org/10.1016/S1567-7443(05)80017-1

Broome MR, Peterson ME (2019). Thyroid imaging. In: EC Feldman, F Fracassi, ME Peterson, eds. Feline Endocrinology. Milan, Italy: EDRA;: 169- 197.

Cao Y, Zhao X, You R, Zhang Y, Qu C, Huang Y, Yu Y, Gong Y, Cong T, Zhao E (2022). CD11c+ B Cells Participate in the Pathogenesis of Graves' Disease by Secreting Thyroid Autoantibodies and Cytokines. Front. Immunol., 13:836347.

Chan A, Carter J (2010). Therapeutic antibodies for autoimmunity and inflammation. Nat. Rev. Immunol., 10: 301-316. https://doi.org/10.1038/nri2761

Cooney M, Vartiainen E, Laatikainen T, De Bacquer D, McGorrian C, Dudina A, Graham I (2012). Cardiovascular risk age: concepts and practicalities. Heart., 98(12): 941-946. https://doi.org/10.1136/heartjnl-2011-301478

Cruse JM, Lewis RE, Wang H (2004). Cluster of Differentiation (Cd) Antigens. Immunol. Guidebook,:47–124. https://doi.org/10.1016/B978-012198382-6/50027-3

Current state and a glycoinformatics perspective. Int. J. Molecul. Sci., 23(9): 5180. https://doi.org/10.3390/ijms23095180

Daniels MA, Devine L, Miller JD, Moser JM, Lukacher AE, Altman JD, Kavathas P, Hogquist KA, Jameson SC (2001). CD8 binding to MHC class I molecules is influenced by T cell maturation and glycosylation. Immunity., 15(6):1051-61. https://doi.org/10.1016/s1074-7613(01)00252-7

De Leo S, Lee SY, Braverman LE (2016). Hyperthyroidism. Lancet, 388(10047):906-918. https://doi.org/10.1016/S0140-6736(16)00278-6

de Oliveira JTS, Wenceslau AA, da Silva EB, de Lavor MSL, Guedes PEB, Carlos RSA (2022). Canine Hypothyroidism with Neurological Disorders. Acta Scient. Vet., 50(1): 808. https://doi.org/10.22456/1679-9216.123247

De Vito P, Incerpi S, Pedersen J, Luly P, Davis F, Davis P (2011). Thyroid hormones asmodulators of immune activities at the cellular level. Thyroid., 21(8): 879-890.

Dev N, Sahoo R, Kulshreshtha B, Gadpayle A, Sharma S (2015). Prevalence of thyroid dysfunction and its correlation with CD4 count in newly-diagnosed HIV-positive adults--a cross-sectional study. Int. J. STD AIDS., 26(13): 965–970. https://doi.org/10.1177/0956462414563776

Duntas LH (2006). The role of selenium in thyroid autoimmunity and cancer. Thyroid., 16(5): 455-460. https://doi.org/10.1089/thy.2006.16.455

Duntas LH (2006). The role of selenium in thyroid autoimmunity and cancer. Thyroid., 16: 455-460.

Eid, S, El-Zaher H, Emara S, Farid O, Michael M (2019). Nano selenium treatment effects on thyroid and selenium: a review. Curr. Med. Chem., 14:1539-1549.

El-Deib M, El-Sharkawy N, Beheiry R, Abd-Elhakim Y, Ismail A, Fahmy E, Saber T (2021). Boldenone undecylenate disrupts the immune system and induces autoimmune clinical hypothyroidism in rats: Vitamin C ameliorative

effects. Int. Immunopharmacol., 99:107939. https://doi.org/10.1016/j.intimp.2021.107939

Ellegård L, Krantz E, Trimpou P, Landin-Wilhelmsen K (2021). Health-related quality of life in hypothyroidism—A population-based study, the WHO MONICA Project. Clin. Endocrinol., 95(1):197-208.

Fakri, Al-Jowari (2023). Antithyroid Peroxidase and Thyroid Hormones in a Sample of Iraqi Patients with Type 2 Diabetic Mellitus. Iraqi J. Sci., 64(4): 1618-1624. https://doi.org/10.24996/ijs.2023.64.4.5

Farhat S, Fatima S, Iqbal M, Arain F (2023). Effects of maternal subclinical hypothyroidism on neurodevelopment of offspring-An animal-based study. Res. Square (Preprint), https://doi.org/10.21203/rs.3.rs-2668084/v1

Felicetti F, Catalano MG, Fortunati N (2017). Thyroid autoimmunity and cancer. Endocr. Immunol., 48:97-109.

Ferrara SJ, Chaudhary P, DeBell MJ, Marracci G, Miller H, Calkins E, Pocius E, Napier BA, Emery B, Bourdette D (2022). TREM2 is thyroid hormone regulated making the TREM2 pathway druggable with ligands for thyroid hormone receptor. Cell Chem. Biol., 29(2):239-248.

Flevaris K, Kontoravdi C (2022). Immunoglobulin G n-glycan biomarkers for autoimmune diseases: diseases. Postepy. Hig. Med. Dosw. (online); 69: 80–90. https://doi.org/10.5604/17322693.1136383

Gauthier B, Sola-García A, Cáliz-Molina M, Lorenzo P, Cobo-Vuilleumier N, Capilla-González V, Martin-Montalvo A (2020). Thyroid hormones in diabetes, cancer, and aging. Aging cell., 19(11): e1326. https://doi.org/10.1111/acel.13260

Ghazi M, Al-Qaiym M (2023). Role of T. vulgaris in protection DNA damage induced by Lead acetate in Rats. J. Biotechnol. Res. Center., 17(1): 55-65. https://doi.org/10.24126/jobrc.2023.17.1.701

Goncharova O (2020). Associative relationships between vitamin D levels, immunological characteristics and thyroid volume in patients with hypothyroidism due to autoimmune thyroiditis. Int. J. Endocrinol. (Ukraine), 16:. 111-116.

Gurda BL, Bradbury AM, Vite CH (2017). Canine and feline models of human genetic diseases and their contributions to advancing clinical therapies. Yale J. Biol. Med., 90(3): 417-431.

Hariharan H, Al-Harbi N, Karuppiah P, Rajaram S (2012). Microbial synthesis of selenium nanocomposite using Saccharomyces cerevisiae and its antimicrobial activity against pathogens causing nosocomial infection. Chalcogenide Lett., 9(12):509-515. https://doi.org/https://chalcogen.ro/509_Hariharan.pdf

Hassan A, Ahmed S (2021). Effectiveness of an Educational Program on Nurses' Knowledge concerning Early Detection of Hypothyroidism/Hyperthyroidism in Baghdad Teaching Hospitals. Pakistan J. Med. Health Sci., 15(3):782-784.

Hong Y, Kim J, Park S, Yi S, Lim M, Lee S, Kang Y(2023). Single cell analysis of human thyroid reveals the transcriptional signatures of aging. Endocrinology., 164(4): bqad029.

Hypothyroidism-associated immunosuppression involves induction of galectin-1-producing regulatory T cells. FASEB J., 37(4):e22865.

Ihnatowicz P, Drywień M, Wątor P, Wojsiat J (2020). The importance of nutritional factors and dietary management of Hashimoto's thyroiditis. Ann. Agricult. Environ. Med., 27(2): 184-193.

Indikar M, Shoukat A(2019). Human Immunodeficiency Virus and Its Co-Relation with Thyroid Levels and Cd4.. OSR J. Dental Med. Sci., 18(5):41-46. https://doi.org/10.9790/0853-1805084146.

Janyga S, Marek B, Kajdaniuk D, Ogrodowczyk-Bobik M, Urbanek A, Bułdak Ł (2021). CD4+ cells in autoimmune thyroid disease. Endokrynologia Polska, 72(5): 572-583.

Kawicka A, Regulska-Ilow B (2015). Metabolic disorders and nutritional status in autoimmune thyroid hormones, immunity and antioxidant status in rabbits. World Rabbit Sci., 27(2): 93-100.

Lagattuta K, Kang J, Nathan A, Pauken K, Jonsson A, Rao A, Sharpe A, Ishigaki K, Raychaudhuri S (2022). Repertoire Analyses Reveal T Cell Antigen Receptor Sequence Features That Influence T Cell Fate. Nat. Immunol., 23: 446–457.

Lei H, Grinberg O, Nwaigwe CI, Hou HG, Williams H, Swartz HM, & Dunn JF (2001). The effects of ketamine–xylazine anesthesia on cerebral blood flow and oxygenation observed using nuclear magnetic resonance perfusion imaging and electron paramagnetic resonance oximetry. Brain Res., 913: 174-179.

Lewiński A, Sewerynek E, Karbownik M (2006). Aging and Age-Related Diseases: The Basics (Aging processes and the thyroid gland). New York: Nova Science Publishers, Inc., 131-172.

LiY, Zhao C, Zhao K, Yu N, Li Y, Yu Y, Zhang Y, Song Z, Huang Y, Lu G, Gao Y, Zhang J, Guo X (2021). Glycosylation of Anti-Thyroglobulin IgG1 and IgG4 Subclasses in Thyroid Diseases. Euro. Thyroid J., 10(2): 114–124. https://doi.org/10.1159/000507699

Liu T, Yang T, Pan T (2018). Effect of Low-Selenium/High-Fat Diet on Pig Peripheral Blood Lymphocytes: Perspectives from Selenoproteins, Heat Shock Proteins, and Cytokines. Biol. Trace Elem. Res., 183:102–113. https://doi.org/10.1007/s12011-017-1122-z

Madhusudhan U, Kalpana M, Singaravelu V, Ganji V, John N, Gaur A, Ganji V (2022). Brain-derived neurotrophic factor-mediated cognitive impairment in hypothyroidism. Cureus, 14(4): e23722. https://doi.org/10.7759/cureus.23722

Maecker HT, McCoy JP, Nussenblatt R (2012). Standardizing immunophenotyping for the human immunology project. Nature Rev. Immuno., 12(3):191-200.

Mangalam A, Rodriguez M, David C(2006). Role of MHC class II expressing CD4+ T cells in proteolipid protein91–110-induced EAE in HLA-DR3 transgenic mice. Euro. J. Immunol., 36(12): 3356-3370.

Mansourian AR (2010). The immune system which adversely alter thyroid functions: a review on the concept of autoimmunity. Pakistan J. Biolog. Sci., 13(16):765-774.

Mathur M, Gupta P, Chaurasia A, Prakash G, Tripathi C (2021). Thyroid Function as Surrogate Marker of Advancing HIV Infection and its Correlation with CD4 Count. Int. J. Sci. Res., 10(6): 461-466. https://doi.org/10.21275/MR21605124907

Mekova RV, Lesichkova SS, Tsakova AD, Hristova JZ, Bakalov DI, Boyanov MA (2020). Circulating CD3(+) CD4(+) CD28((– T lymphocytes in patients with autoimmune. Comptes rendus de l'Académie bulgare des Sciences, 73(5):712-721.

Meng Z, Liu M, Zhang Q, Liu L, Song K, Tan J (2015). Gender and age impacts on the association between thyroid function and metabolic syndrome in Chinese. Medicine, 94(50): e2193.

Mitrou P, Raptis S, Dimitriadis G (2011). Thyroid disease in older

people. Maturitas, 70(1): 5–9. https://doi.org/10.1016/j.maturitas.2011.05.016

Mitrou P, Raptis SA, & Dimitriadis G (2011). Thyroid disease in older people. Maturitas, 70: 5-9.

Mohan B, Damodar K(2019). Functional features of neutrophils in subclinical hypothyroidism compared to euthyroid status. Ann. Med. Physiol., 3(1): 3-9. https://doi.org/10.23921/amp.2019v3i1.19251

Mohan B, & Damodar KS (2019). Functional features of neutrophils in subclinical hypothyroidism compared to euthyroid status: Functional features of neutrophils in subclinical hypothyroidism. Ann. Med. Physiol., 3: 3-9.

Mohsen R, Al-azzawi R, Ad'hiah A(2020). Serum level of interleukin-35 in patients with chronic hepatitis B virus infection. Iraqi J. Sci., 61(11): 2860-2865.

Montesinos MdM, Pellizas CG (2019). Thyroid hormone action on innate immunity. Front. Endocrinol., 10:350. https://www.frontiersin.org/articles/10.3389/fendo.2019.00350

Paulazo M, Klecha A, Sterle H, Valli E, Torti H, Cayrol F, Cremaschi G (2019). Hypothyroidism-related zinc deficiency leads to suppression of T lymphocyte activity. Endocrine., 66: 266-277.

Poe JC, Minard-Colin V, Kountikov EI, Haas KM, Tedder TF (2012). A c-Myc and surface CD19 signaling amplification

Ramezani M, Reisian M, Hezaveh ZS (2023). The effect of synbiotic supplementation on hypothyroidism: A randomized double-blind placebo controlled clinical trial. Plos One., 18(2): e0277213. https://doi.org/10.1371/journal.pone.0277213.

Rustam SS, Hassan BF (2021). Assessment of inflammatory marker (Hs-CRP and OPN) and other parameters in Hypothyroidism. Teikyo Med. J., 44(6):2325-2331.

Sabea A, Alqayim M (2023). Biogenic Selenium Nanoparticles Synergizes Levothyroxine in the Treatment of Hypothyroidism at the Level of Genes Expression. Eur. Chem. Bull. 12(1), 857-872. https://doi.org/10.31838/ecb/2023.12.1.070

Shaji B, Joel J (2022). Impact of Hypothyroidism on Metabolic and Cognitive Dysfunction: A Comprehensive Review. J. Young Pharma., 14(4):349-354. https://doi.org/10.5530/jyp.2022.14.71

Shete A, Thakar M, Abraham P, Paranjape R (2010). A review on peripheral blood CD4+ T lymphocyte counts in healthy adult Indians. Indian J. Med. Res., 132(6); 667-675.

Shihab T, Ibrahim Z (2021). Immunohistochemical and pathological changes in BALB/c mice immunized with whole sonicated Listeria monocytogenes antigens and the effect of probiotics. Iraqi J. Vet. Sci., 35:79-85. https://doi.org/10.33899/IJVS.2021.131562.1970

Shintaro Iwama, Tomoko Kobayashi, Yoshinori Yasuda, Hiroshi Arima (2022), Immune checkpoint inhibitor-related thyroid dysfunction., Best Practice Res. Clin. Endocrinol. Metabol., 36(3):101660.

Silvestri G, Cartwright E (2016). Chapter 9 - HIV and AIDS: Science Wrestles with 10,000 Nucleotides—Points but No Pin. Viral Pathogen., 107-123. https://doi.org/10.1016/B978-0-12-800964-2.00009-4

Stenlund FHK (2022). The diagnosis and treatment of thyroid gland disorders in dogs and cats. Master Thesis: Lithuanian Uni. Health Sci. https://hdl.handle.net/20.500.12512/112787

Sun Y, Teng D, Zhao L, Shi X, Li Y, Shan Z , Teng W (2022). Impaired Sensitivity to Thyroid Hormones Is Associated with Hyperuricemia, Obesity, and Cardiovascular Disease Risk in Subjects with Subclinical Hypothyroidism. Thyroid., 32(4):376–384. https://doi.org/10.1089/thy.2021.0500

Suzuki M, Konya C, Goronzy J, Weyand CM (2008). Inhibitory CD8+ T cells in autoimmune disease. Hum. Immunol., 69(11):781–789. https://doi.org/10.1016/j.humimm.2008.08.283

Tan T, Rajeswaran H, Haddad S, Shahi A, Parvizi J (2016). Increased risk of periprosthetic joint infections in patients with hypothyroidism undergoing total joint arthroplasty. J. Arthropl., 31(4):868-871. https://doi.org/10.1016/j.arth.2015.10.02

Teke Kisa P, Arslan N (2021). Inborn errors of immunity and metabolic disorders: current understanding, diagnosis, and treatment approaches. J. Pediat, Endocrinol. Metabol., 34(3): 277-294.

Valli E, Dalotto-Moreno T, Sterle H, Méndez-Huergo S, Paulazo M, García S, Cremaschi G (2023). Treatment of Hypothyroidism at the Level of Genes Expression, Eur. Chem. Bull., 12(1): 857-872.

Van Poucke M, Van Renterghem E, Peterson M, van den Berg M, Stock E, Peelman L, Daminet S, Venardos K, Kaye D (2007). Myocardial ischemia-reperfusion injury, antioxidant enzyme systems,

Vito P, Incerpi S, Pedersen J, Luly P, Davis F, Davis P (2011). Thyroid Hormones as Modulators of Immune Activities at the Cellular Level. Thyroid., 21(8):879-90. https://doi.org/10.1089/thy.2010.0429

Volpé R. (2001). The Immunomodulatory Effects of Anti-thyroid Drugs are Mediated via Actions on Thyroid Cells, Affecting Thyrocyte-immunocyte Signalling A Review. Curr. Pharmaceut. Design., 7:451-460.

Wali A, Al-Qayim M (2019). Biosynthesis, Characterization and Bioactivity of Selenium Nanoparticles Synthesized by Propolis. Iraqi J. Vet. Med., 43(1): DOI: 10.30539/ijvm

Wang X , Huang J, Zhang A, Fang C, Ma Q, Jiang, P (2021). Altered expression profile of BAFF receptors on peripheral blood B lymphocytes in Graves' disease. BMC Endocr. Disord., 21(8):1-6. https://doi.org/10.1186/s12902-021-00752-3

Wu J, Liang B, Qian Y, Tang L, Xing C, Zhuang Q, Shen Z, Jiang S, Yu K, Feng J (2018). Down-regulation of CD19 expression inhibits proliferation, adhesion, migration and invasion and promotes apoptosis and the efficacy of chemotherapeutic agents and imatinib in SUPB15 cells. Cell Biol. Int., 42(9):1228-1239.

Wu Q, Rayman M, Lv H, Schomburg L, Cui B, Gao C, Chen P, Zhuang G, Zhang Z, Peng X (2015). Low population selenium status is associated with increased prevalence of thyroid disease. J. Clin. Endocrinol. Metabol., 100: 4037-4047.

Wu Z, Guo Z, Zheng Y, Wang Y, Zhang H, Pan H, & Wang W (2023). IgG N-Glycosylation Cardiovascular Age Tracks Cardiovascular Risk Beyond Calendar Age. Engineering (In press). https://doi.org/10.1016/j.eng.2022.12.004

Yan-Jun L, Luo-Ji E, Yu-Qin L (2023). Effect and mechanism of alcohol extract of Alisma orientalis on the thyroid function of mother rats and neurological function of offspring with subclinical hypothyroidism. Med. J. Chinese People's Liberat. Army,. 48(2): 183-189.