Sys Rev Pharm 2020;11(8):706-712
A multifaceted review journal in the field of pharmacy

Thyroid Autoantibodies as a Useful Guideline in Hyperthyroidism Patients Treated with Radioactive Iodine

Zena A. Khalaf [1]*, Hameed M. Jasim [1], Satar M. Kadam [2], Ali A. Mahdi [3]

[1]College of Biotechnology/ Al-Nahrain University, Iraq
[2]College of Medicine, University of Baghdad, Iraq
[3]College of Medical and Health Technology, Middle Technical University, Iraq.
***Corresponding Authors:** zena.shujairi@gmail.com**

ABSTRACT

Background: Several investigations are commonly used in the clinical practice to investigate a patient with hyperthyroidism, including measurement of thyroid autoantibodies (TPO-Ab, TG-Ab, ANA, and dsDNA antibodies).
Methods: Fifty-two hyperthyroidism patients (Graves' disease and Toxic multinodular goiter) and 21 normal healthy volunteers were included in this study. Thyroid antibodies were measured before and after one and four months of radioactive iodine (RAI-131). Treatment success and recovery of patients administer either 10mCi or 20mCi of RAI-131 was analyzed according to clinical data.
Results: A significantly increase in serum levels of TPO-Ab, TG-Ab, ANA antibodies were detected in hyperthyroidism patients before treatment with RAI-131, while a normal levels of dsDSA antibodies was noticed in those patients compared with normal health controls. Serum levels of TPO-Ab, TG-Ab, and ANA antibodies were increased after one month of treatment with RAI-131 (either dose 10 or 20mCi). After four months of radiotherapy, levels of TPO-Ab and TG-Ab antibodies were still increased except a decrease in their levels in GD patient administered the dose 20mCi, while the levels of ANA antibodies were decreased significantly in hyperthyroidism patients treated with RAI-131 except an increase in TMNG patients administered dose 20mCi. On the other hand, serum levels of double-stranded DNA (dsDSA) antibodies were increased significantly in GD patients administered either dose 10 or 20mCi of radioiodine, and TMNG patients administered dose 20mCi.
Conclusion: Successful RAI-131 therapy for hyperthyroidism with 10-20mCi dose can be achieved with decline in thyroid antibodies, ANA and dsDSA.

Keywords: Thyroid autoantibodies, hyperthyroidism, radioactive iodine

Correspondence:
Zena A. Khalaf
College of Biotechnology/ Al-Nahrain University, Iraq
Email: zena.shujairi@gmail.com

INTRODUCTION

Thyrotoxicosis is a clinical state resulting from inappropriately high thyroid hormone levels (1). Endogenous hyperthyroidism is most commonly due to GD or nodular thyroid disease. GD is an autoimmune disorder in which thyrotropin receptor antibodies (TRAb) stimulate the TSH receptor, increasing thyroid hormone production and release. The development of nodular thyroid disease includes growth of established nodules, new nodule formation, and development of autonomy over time (2). Although toxic nodular goiter is less common than GD, its prevalence increases with age and in the presence of dietary iodine deficiency. Therefore, toxic nodular goiter may actually be more common than GD in older patients, especially in regions of iodine deficiency (3,4). Hyperthyroidism is a common problem in pediatric endocrine practices, and it responds well to anti-thyroid medication, it affects 0.5-2% of the population (5). Thyroid nodules, whether benign or malignant often have no symptoms (6).

Histologically, there is lymphocytic infiltration in the thyroid tissue and autoreactive response is directed against thyroid autoantigens; as well as production of specific autoantibodies, which can be directed against thyroid peroxidase antibodies (TPO-Ab), thyroglobulin, and/or thyroid stimulating hormone receptor (7). The TPO antibody is more sensitive marker for autoimmune thyroid dysfunction than Tg (thyroglobulin) antibody (8). It was suggested that the TPO antibody titers are associated with the extent of lymphocytic infiltration of thyroid tissue (9). Autoimmune thyroid diseases are usually accompanied by the presence of thyroid peroxidase antibody (TPO), thyroglobulin antibody (Tg), and thyroid stimulating hormone receptor (TSHR) antibodies (10), In addition to anti-nuclear antibodies and double strand antibodies which attack thyroid gland and destruct tissues. Antinuclear antibodies (ANA) are recognized as valuable diagnostic marker in evaluating systemic autoimmune diseases (e.g., systemic lupus erythematosus and mixed connective tissue disorder) (11). Double strand DNA antibodies have different subclasses, including IgA, IgE, IgG, and IgM. Double strand DNA antibody originated from Nuclear antigens, such as dsDNA, which are not accessible to the immune system because they are restricted to the nucleus and mitochondria and are quickly degraded by DNases in the cytoplasm and endosomes (12, 13,14).

Hypothyroidism radiotherapy may be influenced by many factors, such as RAI-131 dosage, age, gender, size of the thyroid gland, initial FT4, FT3, TSH levels, radioactive iodine uptake, duration of disease, administration of antithyroid drugs and the presence of thyroid antibodies (15). The damage of thyroid gland RAI-131 therapy leads to an immunological response that may be disadvantageous or beneficial (16). In this study, the dynamic changes of serum thyroid peroxidase antibody (TPO-Ab), Thyroglobulin antibody (TG-Ab), Double strand DNA ab (dsDSA), anti-Nuclear antibody (ANA) in Graves' disease (GD) and TMNG patients were analyzed before and after radioactive iodine threapy to investigate if autoantibodies play a pivotal role in the occurrence of

early hypothyroidism and after RAI-131 treatment of hyperthyroidism patients.

MATERIALS AND METHODS

A total of 52 hyperthyroidism patients and 21 normal healthy controls were enrolled in this study. Blood samples were collected from patients diagnosed as hyperthyroidism (Graves and Toxic multinodular goiter) attends Baghdad center for radiotherapy/Nuclear Medicine Hospital in Baghdad Governorate during the period from January/2018 to September/2019. Hyperthyroidism patients were diagnosed by clinical examination of nuclear thyroid scan and or thyroid ultrasound in hospital radiation unit in addition to biochemical tests before treatment to identify the etiology of hyperthyroidism, size of thyroid gland and radiation dose for each patient. Patients administered RAI-131 (10 or 20 mCi) were followed up after treatment for one and four months to evaluate the effect of radiotherapy on the levels of autoantibodies. Ethical approval for this study was obtained from the Ministry of health, the directorate of health in Al-Resafa sector/Baghdad Governorate.

Serum assay

Blood samples were collected from patients before radioiodine therapy and after 1 and 4 months of treatment. Serum circulating TSH, TPO-Ab, TG-Ab, ANA and dsDNA were measured by ELISA analyzer using special auxiliary reagents (17; 18; 19; 20 and 21 respectively). Serum TR-Ab was measured by radiation receptor method using assay kit (Calbiotech, USA). The recovery rate was determined according to serum concentrations of TSH after radioiodine therapy.

Statistical analysis

Statistical analysis was performed with SPSS software version 19.0. Data was presented as means ± standard deviation. One-way ANOVA was used to detect multivariate significance ($P < 0.01$, $P < 0.05$). t test was used to compare significant differences between groups.

RESULTS AND DISCUSSION

Effect of RAI-131 on Thyroid peroxidase antibodies

Thyroid peroxidase antibodies (TPO-Ab) were used as a prognostic indicator biomarker for detection of thyroid dysfunction. Results indicated in figure 1 showed that serum levels of TPO was significantly increased ($P \leq 0.01$) in GD and TMNG patients compared with healthy controls, with higher levels in GD patients than TMNG patients ($P \leq 0.01$). The significant increase may be due to autoimmune response to production of high levels of thyroid autoantibodies (17). TPO-Ab is an important thyroid autoantibody which is commonly found in patients with thyroid diseases, and it correlate with the severity of lymphocytic infiltration and could induce antibody-dependent cell-mediated cytotoxicity (18).

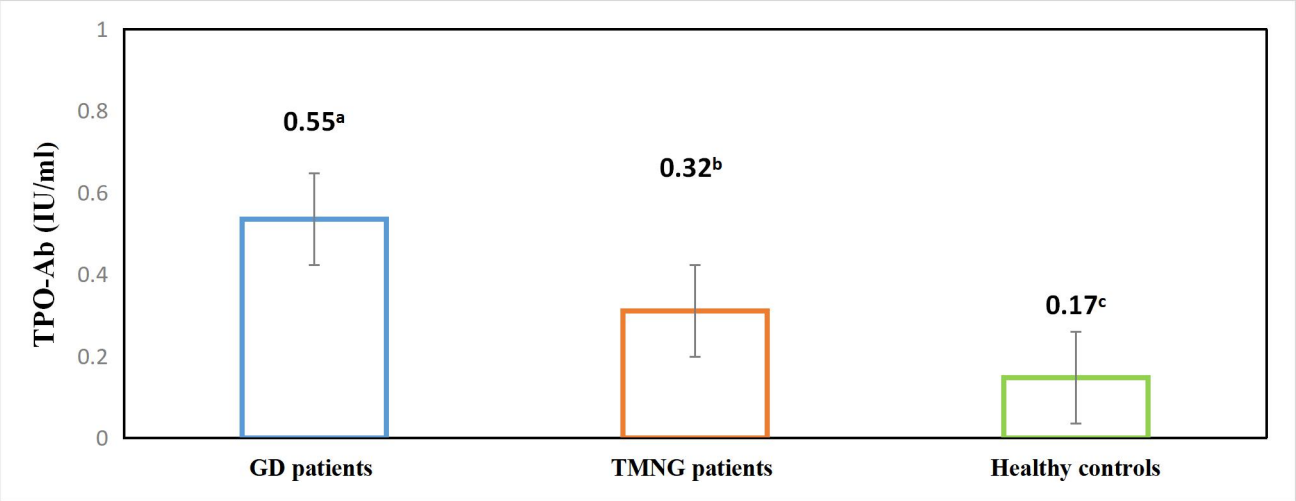

Figure 1: Serum levels of Thyroid peroxidase antibodies in hyperthyroidism patients. Different letters on columns indicates a significant difference ($P \leq 0.01$).

On the other hand, Results indicated in table 1 showed that serum levels of TPO-Ab were significantly increased ($P \leq 0.01$) in GD patients after one and four months of RAI-131 treatment with with dose10mCi, while it showed that there are no significant differences in TPO-Ab levels of GD patients after one month of treatment with dose 20mCi, then the levels were significantly decreased ($P \leq 0.01$) after four months of treatment. These results may be explained by the fact that TPO-Ab is a good marker for individuals with autoimmune hypothyroidism after radiotherapy who have high levels of TSH or less than normal T3 or T4.

Table 1: Serum levels of Thyroid peroxidase antibodies (TPO-Ab) in hyperthyroidism patients before and after treatment with RAI-131

Disease	RAI dose (mCi)	Thyroid peroxidase antibodies (IU/ml) ±SD		
		Before	After Month 1	After Month 4
GD	10	$0.500^{a} \pm 0.12$	$0.817^{b} \pm 0.12$	$0.921^{c} \pm 0.22$

	20	$0.496^{a} \pm 0.16$	$0.596^{b} \pm 0.37$	$0.417^{b} \pm 0.39$
TMNG	10	$0.419^{a} \pm 0.16$	$0.514^{b} \pm 0.12$	$0.577^{b} \pm 0.03$
	20	$0.203^{a} \pm 0.04$	$0.311^{b} \pm 0.04$	$0.395^{b} \pm 0.38$

Different letters in rows indicates a significant difference (P≤0.01), SD: Standard deviation

Furthermore, a significant decrease (P≤0.01) in TPO-Ab levels was observed after one month of treatment with RAI-131 in patients developing hypothyroidism or euthyroidism but not in those remaining hyperthyroid. Because both the TSH receptor and thyroid peroxidase are membrane proteins, the post radioiodine increases serum levels of the correspondent autoantibodies that may be regarded as a marker for thyroid cell damage. Conceivably, this phenomenon is related to the degree of thyroid damage after RAI-131 treatment, to the subsequent reduction in thyroid volume, and the development of hypothyroidism (19). These findings suggest that positive TPO-Ab titers and the presence of smaller goiters may increase the risk for the development of hypothyroidism (22)

A high level of TPO-Ab was positively associated with the risk of thyroid nodules. Results indicated in table 1 showed that there are a significant difference in TPO-Ab levels after one month of treatment in TMNG patients treated with RAI-131 (10 and 20mCi), as the TPO-Ab was significantly increased (P≤0.01) after one month of treatment. While these levels were non significantly increased (P≤0.01) after four months of treatment of TMNG patients with high and low dose (10 and 20mCi) as the levels of TPO-Ab in serum samples of those patients after four months was not statically different than their levels after one month of treatment as shown in table 1. These findings are similar to the results obtained by (20) who mentioned that 5% of toxic goiter (adenoma or multinodular goiter) developed thyroid peroxidase antibodies after one and three months of radioiodine therapy and the percentage was increased up to 22% in GD patients. Positivity for TPO-Ab antibodies was a risk factor for developing hypothyroidism after radioactive iodine treatment (24).

Effect of RAI-131 on Thyroglobulin antibodies

Thyroglobulin (TG) is antigen largely recognized in the sera of patients with autoimmune thyroid disorders (23). Results indicated in figure 2 showed that serum levels of TG-Ab in GD patients were significantly higher (P≤0.01) than their levels in TMNG patients and healthy controls. However, in regard to TMNG patients, TG-Ab and TPO-Ab are two important thyroid autoantibodies to assess the risk of thyroid nodules, a high level of TG-Ab was positively associated with risk of thyroid nodules (24). The existence of thyroid autoimmunity (positive TG-Ab or TPO-Ab) in TMNG patients at the time of RAI-131 treatment is another risk factor for Graves' disease that induced by high-dose internal irradiation (25).

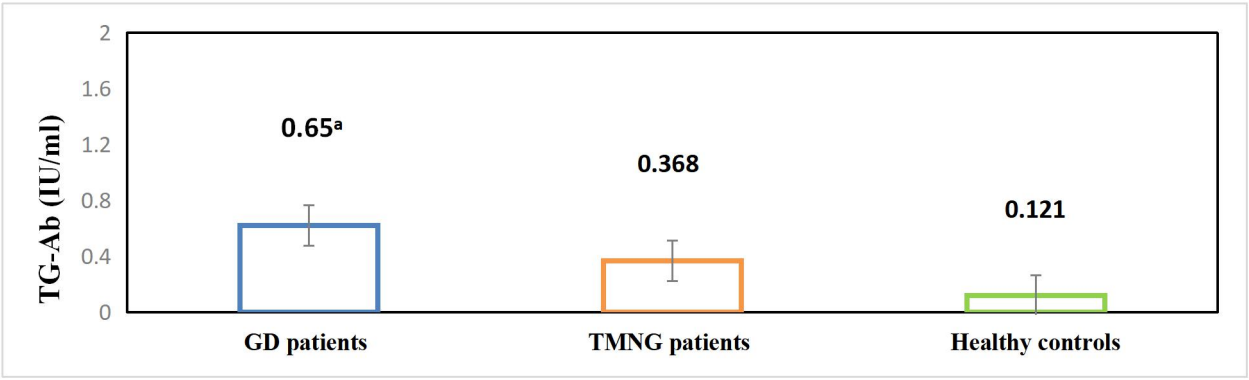

Figure 2: Serum levels of thyroglobulin antibodies in hyperthyroidism patients before treatment with RAI-131. Different letters on columns indicates a significant difference (P≤0.01).

Results indicated in table 2 showed that the serum level of TG-Ab in GD patients treated with RAI-131 dose 10mCi and TMNG patients treated with high and low dose of RAI-131 (10 and 20mCi), was increased significantly (P≤0.01) after one and four months of treatment, while there is no significant difference in serum TG-Ab in GD patients before and after treatment with a high dose of RAI-131 (20mCi), then the level of serum TG-Ab was decreased significantly (P≤0.01) after four months of treatment. The post radioiodine increase in serum levels of the correspondent autoantibodies may be regarded as a marker of thyroid cell damage produced by RAI-131, this is related to the degree of thyroid damage produced by RAI-131, to the subsequent reduction in thyroid volume, and the development of hypothyroidism (19).

Table 2: Serum levels of thyroglobulin antibodies in hyperthyroidism patients before and after treatment with RAI-131.

Disease	RAI (mCi)	Thyroglobulin antibodies (IU/ml)	P-value

		Before	After Month 1	After Month 4	
GD	10	0.541[a] ± 0.12	0.914[b] ± 0.12	0.931[b] ± 0.16	** 0.0001
	20	0.705[a] ± 0.31	0.732[a] ± 0.56	0.662[b] ± 0.35	** 0.0001
TMNG	10	0.376[a] ± 0.30	0.479[b] ± 0.24	0.509[c] ± 0.16	** 0.0001
	20	0.363[a] ± 0.12	0.545[b] ± 0.04	0.572[b] ± 0.16	** 0.0092

Different letters in rows indicates a significant difference (P≤0.01), SD: Standard deviation

These results are similar to (26) who revealed that as for thyroglobulin antibodies (TGAb), the response seems biphasic with an initial rapid decline and a subsequent increase above baseline levels after radioactive iodine treatment.

Effect of RAI-131 on Anti-Nuclear autoantibodies

Antinuclear antibodies (ANAs), are antibodies against specific non–thyroid antigens, have been described in patients with autoimmune thyroid disease (AITD), whereas the prevalence of ANA varies from 9-35% and is much higher when TPO-Ab and TG-Ab are presents in high levels (75% and 69%, respectively) (27). In this study, we detect a correct interpretation of the presence of ANAs in immune and non-immune hyperthyroidism patients and investigate the correlation between radioactive iodine response and recovery outcomes with ANA prevalence in GD and TMNG patients. Results indicated in figure 3 showed that serum level of ANA in GD and TMNG patients was significantly higher (P≤0.01) than their levels in healthy controls before radiotherapy.

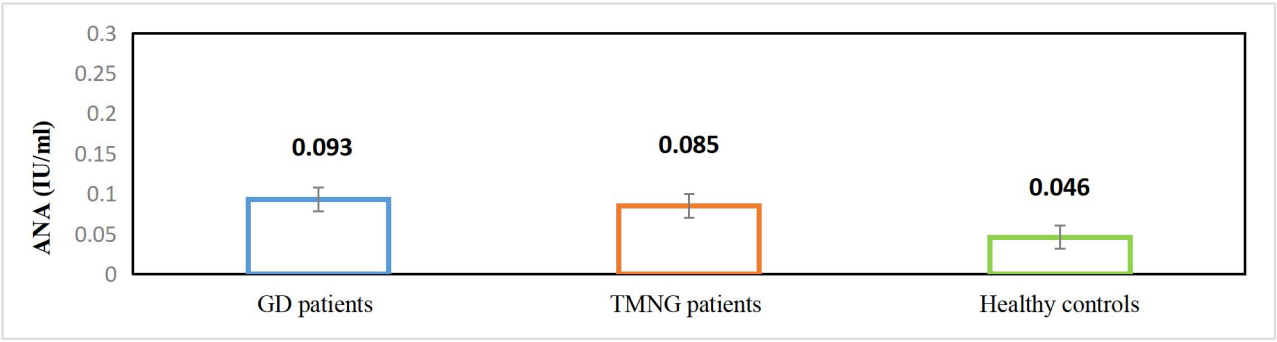

Figure 3: Serum levels of antinuclear antibodies in hyperthyroidism patients before treatment with RAI-131. Different letters on columns indicates a significant difference (P≤0.01).

Results indicated in figure 3 also showed that there is no significant difference in serum levels of ANA in GD and TMNG patients before RAI-131 treatment. Furthermore, ANA levels in hyperthyroidism patients are significantly higher (P≤0.01) than their levels in healthy controls before radioiodine therapy. On the other hands, results indicated in table 3 showed that serum levels of ANA were significantly increased (P≤0.01) after one month of treatment with RAI-131 (10 and 20mCi) in all GD and TMNG patients. While After four months of radiotherapy, it was found that levels of ANA were significantly decreased (P≤0.01) in GD and TMNG patients treated with low dose of RAI-131 (10mCi) which means that low dose of RAI-131 may have short term effect on ANA in patients. Results also showed that serum levels of ANA in GD patients treated with 20mCi was increased significantly (P≤0.01) after one month of treatment then decreased significantly after four months of treatment, this may be due to delayed response to RAI-131 and their role in persistent hyperthyroidism as mentioned by (28) who reported that autoimmune hyperthyroidism patients had a positive frequency of ANA (60%), TPO-Ab (21%) and TG-Ab (29%) and the positivity of ANA was an indicator of increased risk of developing hypothyroidism overtime after RAI-131 treatment. Furthermore, results indicated in table 2 showed that the serum level of ANA in TMNG patients had no significant difference than their levels effect after four months of treatment with 20mCi.

Table 3: Serum level of antinuclear antibodies in hyperthyroidism patients before and after RAI-131 treatment

Disease	RAI (mCi)	Antinuclear Antibodies (IU/ml) ±SD			P-value
		Before	After Month 1	After Month 4	
GD	10	0.095[a] ± 0.01	0.193[b] ± 0.008	0.060[c] ± 0.01	0.0001**

	20	$0.091^{a} \pm 0.01$	$0.155^{b} \pm 0.07$	$0.087^{c} \pm 0.05$	0.0001**
TMNG	10	$0.091^{a} \pm 0.02$	$0.117^{b} \pm 0.01$	$0.061^{c} \pm 0.004$	0.0057**
	20	$0.082^{a} \pm 0.01$	$0.168^{b} \pm 0.01$ b	$0.108^{b} \pm 0.03$	0.0327*

Different letters in rows indicate a significant difference **: (P≤0.01); *: (P≤0.05), SD: Standard deviation

In this study, it was found that there is a high association between serum levels of ANA and the recovery rate in all hyperthyroidism patients which was the first findings in this field been there are rare studies about the correlation between RAI-131 response and its effect with ANA in thyroid disorders, according to their results, it was found that there is a positive correlation between ANA and immune and non-immune hyperthyroidism patients.

Effect of RAI-131 on Double stranded DNA antibodies

A high prevalence of antibodies to double-stranded DNA (dsDSA) antibodies has been recently reported in serum of patients with autoimmune thyroid disorders. In this study, the prevalence of these antibodies has been evaluated in hyperthyroidism patients. Results illustrated in figure 4 showed that there is no significant difference between serum levels of dsDSA antibodies in GD and TMNG patients, and healthy controls before radioiodine treatment.

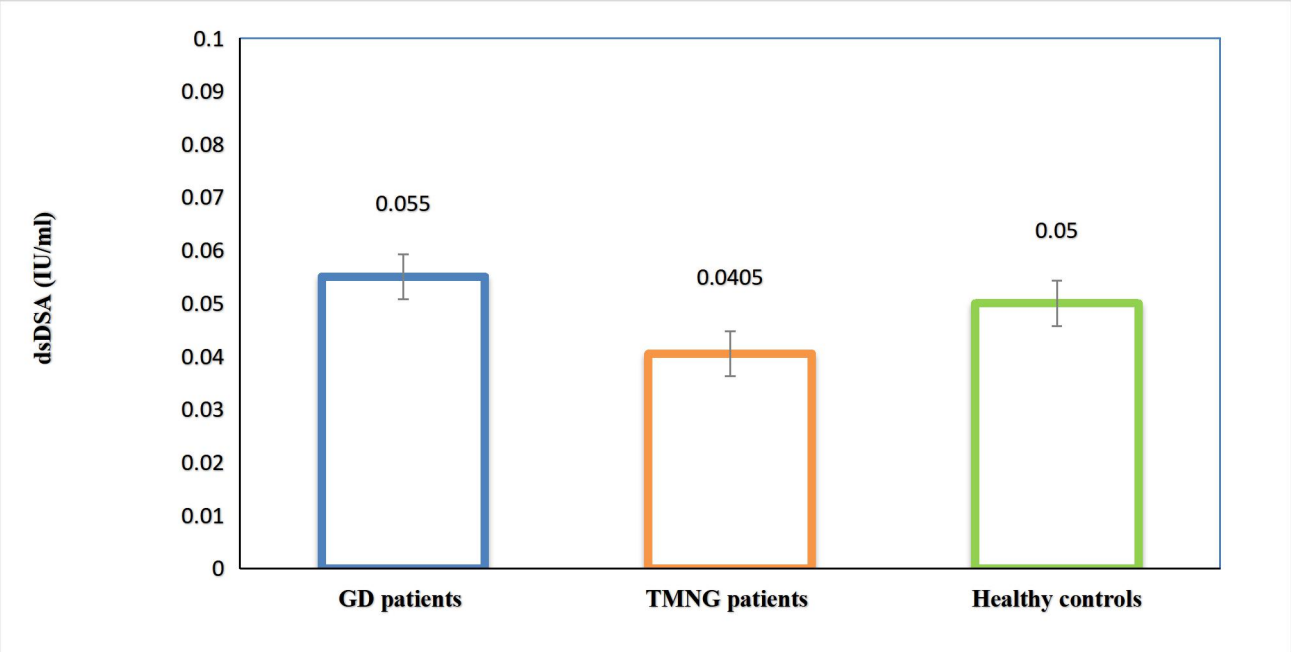

Figure 4: Serum levels of double stranded DNA antibodies in hyperthyroidism patients before treatment with RAI-131. Different letters on columns indicates a significant difference (P≤0.01).

These novel results confirmed that there is no correlation between dsDNA antibodies levels and hyperthyroidism, while single-stranded DNA antibodies were previously reported even in healthy people, but the rate of positivity was very low (29). Results indicated in table 3 showed that there are no significant differences in serum levels of dsDNA antibodies before and after treatment of GD patients with low dose of RAI-131 (10mCi), while it was found that there is a significant increase in their levels in GD patients treated with a high dose of RAI-131 (20mCi) showed a highly significant increase after one and four months of treatment respectively.

Table 4: Serum levels of double-stranded antibodies in hyperthyroidism patients before and after RAI-131

Disease	RAI (mCi)	Double strand antibodies (IU/ml) ±SD			P-value
		Before	After Month 1	After Month 4	
GD	10	$0.050^{a} \pm 0.007$	$0.031^{a} \pm 0.01$	$0.053^{a} \pm 0.008$	NS 0.791
	20	$0.068^{a} \pm 0.02$	$0.095^{b} \pm 0.05$	$0.255^{c} \pm 0.03$	0.0063**
TMNG	10	$0.040^{a} \pm 0.008$	$0.037^{a} \pm 0.01$	$0.053^{a} \pm 0.02$	NS 0.715

	20	0.041[a] ± 0.01	0.051a ± 0.01	0.097b ± 0.05	0.0452*

Different letters in rows indicate a significant difference ** :(P≤0.01); ** :(P≤0.05), NS: Non-significant, SD: Standard deviation.

The novel findings in this study are that there is a slight correlation between dsDNA level and recovery of hyperthyroidism in TMNG patients. Environmental factors such as drugs, infection, and some vaccines also alter the anti-DNA antibodies response in humans (22).

CONCLUSION

A significant increase in thyroid peroxidase, thyroglobulin, and antinuclear antibodies in GD and TMNG patients before radioiodine therapy compared with healthy controls, while there is no significant difference in dsDSA antibodies among hyperthyroidism patients and healthy controls. Serum levels of TPO-Ab, TG-Ab, and ANA antibodies, were increased after one month of treatment with RAI-131 (either dose 10 or 20mCi), while there are variable levels of these antibodies after four months of radioiodine therapy. The levels of Double strand DNA antibody were non-significant in GD and TMNG patients before treatment compared with healthy controls before radioiodine therapy. Serum levels of ANA showed non-significant difference in GD and TMNG patients treated with low dose of RAI-131 (10mCi), while it showed that dsDSA antibodies levels increased significantly after one and four months of treatment in GD and TMNG patients treated with RAI-131 (20mCi).

REFERENCES

1. Ross, D.S., Burch, H.B., Cooper, D.S., Greenlee, M.C., Laurberg, P., Maia, A.L., et al. (2016). American Thyroid Association Guidelines for Diagnosis and Management of Hyperthyroidism and Other Causes of Thyrotoxicosis. Thyroid, 26, 1343-1421.
2. Berghout, A., Wiersinga, W.M., Smits, N.J. and Touber, J.L. (1990). Interrelationships between Age, Thyroid Volume, Thyroid Nodularity, and Thyroid Function in Patients with Sporadic Nontoxic Goiter. The American Journal of Medicine, 89, 602-608.
3. Laurberg, P., Pedersen, K.M., Hreidarsson, A., Sigfusson, N., Iversen, E. and Knudsen, P.R. (1998). Iodine Intake and the Pattern of Thyroid Disorders: A Comparative Epidemiological Study of Thyroid Abnormalities in the Elderly in Iceland and in Jutland, Denmark. The Journal of Clinical Endocrinology and Metabolism, 83, 765-769.
4. Abraham-Nordling, M., Byström, K., Törring, O., Lantz, M., Berg, G., Calissendorff, J., et al. (2011) Incidence of Hyperthyroidism in Sweden. European Journal of Endocrinology, 165, 899-905.
5. Brown, S., Bremner, A., Hadlow N. et al., (2016).The log TSH-free T4 relationship in a community-based cohort is nonlinear and is influenced by age, smoking and thyroid peroxidase antibody status, Clinical Endocrinology, vol. 85, no. 5, pp. 789–796.
6. Shabsigh, R.; Arver, S.; Channer, K.; Eardley, I.; Fabbri, A. and Gooren (2008). The triad of erectile dysfunction, hypogonadism and the metabolic syndrome. International Journal of Clinical Practice, 62:791–798.
7. Leger, J., Oliver, I., Rodrigue, D, Lambert S, and Coutant R. (2018). Consensus: Graves' disease in children. Annals Endocrinology journal; 79:647–55.
8. Ron, E., and A. Brenner. (2010). Non-Malignant Thyroid Diseases Following a Wide Range of Radiation Exposures. Radiat Res.; 174(6): 877–888. doi:10.1667/RR1953.1.
9. Hasan R., and Raziq A., 2019. Studying the Frequency of Autoimmune Thyroid Diseases in Duhok Province. Science Journal of University of Zakho 7(2), 45-49.
10. Citterio, E., Veluswamy, B., Morgan S., et al. (2017). De novo triiodothyronine formation from thyrocytes activated by Thyroid Stimulating Hormone. J. Biol. Chem. 292, 15434–15444.
11. Ruggeri, M., Vicchio, M., Cristani, M., Certo, R., Caccamo, D., Alibrandi, A., et al. (2016). Oxidative stress and advanced glycation end products in Hashimoto's thyroiditis. Thyroid. 26:504–11.
12. Frohlich, E., and R. Wahl. (2017). Thyroid Autoimmunity: Role of Anti-thyroid Antibodies in Thyroid and Extra-Thyroidal Diseases. Front. Immunol. 8:521.
13. Thushani, S., Karthik, K., Vinodh, R., Vasanth, J., Tianhao, W., et al., (2018). Exploring Systemic Autoimmunity in Thyroid Disease Subjects. Journal of Immunology Research, Volume (2018), Article ID 6895146, 7 pages.
14. Mistry, P., and J. Kaplan (2017). Cell death in the pathogenesis of systemic lupus erythematosus and lupus nephritis. Clin. Immunol. 185:59–73.
15. Hodhod, E. (2013). Thyrotoxicosis: A Retrospective Study of Cases Seen at Nuclear Medicine Unit (Nemrock). PhD Thesis. Cairo University. Faculty Of Medicine. Egypt.
16. Barnado A, Crofford J, and C. Oates. (2016). At the bedside: neutrophil extracellular traps (NETs) as targets for biomarkers and therapies in autoimmune diseases. J. Leukoc. Biol. 99:265–78.
17. Ahmad, T., Khoja, A., Rashid, N. H., and M. A. Ashfaq. (2018). Outcome of radioactive iodine therapy in Toxic Nodular Goiter in Pakistan. Pakistan journal of medical sciences, 34(5), 1146–1151.
18. Ahmad, T., and A. Khoja. (2018). Outcome of radioactive iodine therapy in Toxic Nodular Goiter in Pakistan. Pak. J. Med. Sci., Vol. 34, No.5.
19. Chiovato L., Fiore E., Vitti P., Rocchi R., Rago T., Dokic D., Latrofa F., Mammoli C., Lippi F., and C. Ceccarelli. (1998). Outcome of Thyroid Function in Graves' Patients Treated with Radioiodine: Role of Thyroid-Stimulating and Thyrotropin-Blocking Antibodies and of Radioiodine-Induced Thyroid Damage. J. Clin. Endocrinol. Metab. 83(1):40–46.
20. Nagayama, Y. (2018). Radiation-related thyroid autoimmunity and dysfunction. Journal of radiation research, 59 (suppl_2), 98–107.
21. Ahmad, T., Khoja, A., Rashid, N. H., and M. A. Ashfaq. (2018). Outcome of radioactive iodine therapy in Toxic Nodular Goiter in Pakistan. Pakistan journal of medical sciences, 34(5), 1146–1151.

22. Pavlovic M, Kats A, Cavallo M, Chen R, Hartmann JX, Shoenfeld Y. Pathogenic and Epiphenomenal Anti-DNA Antibodies in SLE. Autoimmune Dis. 2010;2011: 462841. Published 2010 Jul 20. doi:10.4061/2010/462841
23. Freitas, E. (2000). Therapeutic Options in the management of toxic and nontoxic nodular goiter. Semin. Nucl. Med., 30:88-97.
24. Xu, W., Liangliang Huo, Zexin Chen, Yangmei Huang, Xingyi Jin, Jing Deng, Sujuan Zhu and Y. Yunxian. (2017). The Relationship of TPO-Ab and TG-Ab with Risk of Thyroid Nodules: A Large Epidemiological Study. Int. J. Environ. Res. Public Health 2017, 14, 723.
25. Elizabeth, N., Pearce, A. and Lewis E. (2003). Thyroiditis, N. Engl. J. Med; 348: 2646- 55.
26. Steen Joop Bonnema and Laszlo Hegedu¨ s, (2012). Radioiodine Therapy in Benign Thyroid Diseases: Effects, Side Effects, and Factors Affecting Therapeutic Outcome. Endocrine Reviews, 33(6):920–980. edrv.endojournals.org
27. Soy, M., Guldiken, S., Arikan, E., Altun, U., and A. Tugrul. (2007). Frequency of rheumatic diseases in patients with autoimmune thyroid disease. Rheumatol. Int., 27:575–7.
28. Torok, K. and T. Arkachaisri. (2010). Autoimmune thyroiditis in antinuclear antibody positive children without rheumatologic disease. Pediatr. Rheumatol. Online J.; 8: 15.
29. Watanabe, A., Kodera, M., Sugiura, K., Usuda, T., Tan, E., Takasaki, Y., Tomita, Y., and Y. Muro, (2004). Anti-DFS70 antibodies in 597 healthy hospital workers. Arthritis and Rheumatology.50: 3, 112-123.