Trace Element Status and Hypothyroidism: A Systematic Review and Meta-analysis

Talebi, S., Ghaedi, E., Sadeghi, E., Mohammadi, H., Hadi, A., Clark, C. C. T. & Askari, G.

Original citation & hyperlink:
Talebi, S, Ghaedi, E, Sadeghi, E, Mohammadi, H, Hadi, A, Clark, CCT & Askari, G 2020, 'Trace Element Status and Hypothyroidism: A Systematic Review and Meta-analysis', Biological Trace Element Research, vol. 197, no. 1, pp. 1-14.
https://dx.doi.org/10.1007/s12011-019-01963-5

DOI 10.1007/s12011-019-01963-5
ISSN 0163-4984
ESSN 1559-0720

Publisher: Humana Press

Trace element status and hypothyroidism: A systematic review and meta-analysis

Abstract

Background: The metabolism of thyroid hormones has been linked with alterations in trace element levels. However, previous reports comparing trace element levels in hypothyroid patients and healthy individuals had yielded equivocal results. Therefore, the aim of this meta-analysis was to investigate the association between trace element (selenium (Se), zinc (Zn), iron (Fe), manganese (Mn), copper (Cu), magnesium (Mg) and lead (Pb)) concentrations in hypothyroid patients and healthy subjects.

Methods: Electronic databases including PubMed, Scopus, Embase, and Web of Science were searched systematically until October 2018. Twenty-five observational studies were included. Hedges' g was used to represent effect sizes, due to trace element levels being presented in different units among studies.

Results: Our results revealed that lower Se concentration in patients with hypothyroidism compared with controls (Hedges' g= −0.45; 95% CI = [−0.88, -0.02]; P = 0.042). In the subgroup analysis, Zn level was found to be lower in overt hypothyroid patients (Hedges' g= −1.19; 95% CI = [−2.33, -0.04]; P = 0.04), while Fe (Hedges' g= −1.11; 95% CI = [-2.21, -0.02]; P = 0.046) and Se (Hedges' g= −0.51; 95% CI = [-0.86, -0.16]; P = 0.004) levels were significantly lower in subclinical hypothyroidism.

Conclusion: We found lower Se concentration in patients with hypothyroidism. In addition, we found decreased concentration of Zn in overt hypothyroid patients and decreased Fe and Se level in subclinical hypothyroidism. Further studies, with higher quality and larger sample sizes, are required to explicate the link between trace element status and hypothyroidism.

Key Words: Trace elements, Hypothyroidism, Meta-analysis

Introduction

Hypothyroidism is a major endocrine disorder which is defined as a disturbance in levels of circulating thyroid hormones; whilst autoimmune disorders, irradiation, or thyroid resection are considered to be potential etiological triggers [1]. The prevalence of hypothyroidism is reported to be up to two percent in geographical areas with sufficient iodine intake [2]. Thyroid hormones exert several physiological functions in the human body; such as regulation of body temperature, modulation of carbohydrates, lipids and proteins metabolism and also regulation of electrolytes and minerals metabolism [3].

Several trace elements like selenium (Se) and zinc (Zn) are involved in the synthesis and metabolism of thyroid hormones [4]. Furthermore, Se is a cofactor of iodothyronine deiodinase, which converts T_4 to T_3, and plays an imperative role in the regulation of thyroid hormones synthesis. Se conceivably acts as a glutathione reductase and glutathione peroxidase cofactor, which protects the thyroid gland against oxidative stress [5]. Zn is reported to be essential for optimal thyroid function [6], is essential in thyroid hormones conversion [7], and is also known as the cofactor of thyrotropin releasing hormone biosynthetic enzyme [7, 8]. Iron (Fe) enhances the activity of the thyroid peroxidase enzyme, which causes iodine binding to thyroid hormones [9, 10].

Previous reports evaluating the association between the circulating levels of trace element and hypothyroidism have shown conflicting results; with some studies [4, 11, 12] indicating increased circulating levels of trace elements, whilst others [12-15] have reported decreased concentrations. Therefore, the aim of the present meta-analysis was to summarize current

evidence on the circulating levels of Zn, manganese (Mn), copper (Cu), Se, magnesium (Mg), Fe and lead (Pb) levels between hypothyroid patients and healthy human adults.

Methods

Search Strategy

We searched the databases PubMed, Science Direct, Embase and Scopus up to October 29, 2018. Search strategies including the key terms and the queries for each database are presented in the supplementary table 1.

Selection Criteria

Studies were included in the final analysis if they: 1) used case-control or cross-sectional design; 2) conducted investigations on adults cases with hypothyroidism (age > 18 years) and healthy controls; 3) reported at least the mean and standard deviation of one of the trace elements (Zn, Mg, Se, Fe, Cu, Mn, Pb) in serum or plasma levels. Studies were excluded if they: 1) were trials, animal studies, case reports, conference papers, letters, editorial or reviews studies; 2) did not have adequate information to be extracted; 3) conducted on children or pregnant women; 4) were uncontrolled studies or conducted on non-healthy controls; 5) reported the levels of trace elements in erythrocytes. All studies were separately assessed by two investigators to determine adherence to selection criteria.

Data extraction

Data extraction was independently performed by two authors (HM and ST) using the standard form of pre-designed data collection. The following data was extracted from articles: first author's name, year of publication, country, study design, sample size, mean age, proportion of

male in groups, matched variables between the case and control group, body mass index and levothyroxine use.

Quality assessment

Two reviewers (HM and ST) scored the included studies using a Newcastle–Ottawa Scale (NOS) [16], including 3 major domains : selection of participants (0-4 stars), comparability of groups (0-2stars) and ascertainment of outcome (0-3stars). Scores of ≤ 3, 4–6 and ≥ 7 were considered as low, moderate and high quality studies respectively. Any disagreement in quality assessment was resolved through panel discussion.

Statistical Analysis

We calculated the standardized mean difference (SMD) with 95% CI using the Hedges statistic. Random effect model based on Inverse-Variance method was used in STATA (version 13) to pool the data. We assessed and quantified heterogeneity using heterogeneity chi-squared test with P-value less than 0.1 and I^2 statistic over 50% considered as significant heterogeneity among studies. We ran a subgroup analysis for the case category variable (whether cases belong to category 1 or 2). Sensitivity analysis was performed to check whether the results were sensitive to the exclusion of one or more studies. We used Begg's and Egger's tests to check for the publication bias, and finally, trim and fill analysis was performed to check if observed publication bias could affect the results.

Results

Literature search

Figure 1 illustrates the PRISMA flow diagram of studies through the selection process for inclusion in the systematic review and meta-analysis. A total of 818 studies identified from Medline, Embase, Scopus and Web of science databases. Five hundred and forty-one records were screened by title and abstract, and after duplicates were removed, 474 records were excluded. Among 67 remaining studies, 42 articles were excluded by full-text assessment. The most common reasons for exclusion were inappropriate data reporting (n=10), unavailable full texts (n=3), conducted on pregnant women (n=3), children (n=5), goiter (n=2) or hyperthyroid subjects (n=11), and inappropriate control group (n=8). Finally 25 case-control or cross-sectional studies included in the present meta-analysis [3, 4, 11-15, 17-34]. From these studies 11, 10, 9, 8, 6, 4 and 3 articles reported Mg [3, 13, 15, 17, 18, 20, 22, 28-30, 34], Zn [4, 11-15, 20, 23, 27, 29], Se [4, 12, 13, 19-21, 24, 32, 33], Cu [4, 11-14, 20, 23, 29], Fe [4, 11, 20, 25, 26, 28], Mn [4, 11-13] and Pb [4, 11, 29], respectively.

Study characteristics

The characteristics of the included studies are reported in **Table 1**. Included studies were published in numerous countries between 1996 and 2018. The selected studies contained overt hypothyroidism [3, 4, 11-15, 17-19, 22-31, 33, 34], subclinical hypothyroidism [17, 20, 21, 24, 26, 27, 29, 31, 32] and euthyroid [17, 19] diagnosed participants, while all the studies contained a control group of healthy subjects. Most records presented both genders in adults and elderly.

Twenty-three articles [3, 4, 11-15, 17-28, 30-32, 34] entered into meta-analyses presented data on serum levels of trace elements, whereas two articles [29, 33], reported data on plasma levels.

Quality assessment

The results of NOS, to indicate the quality assessment of included articles, is showed in Table 2. Among included studies, 12 records [11, 17, 19, 20, 23, 24, 27, 28, 30-32, 34] were identified as high quality (NOS ≥ 7) and the remaining studies [3, 4, 12-15, 18, 21, 22, 25, 26, 29, 33] were identified as moderate quality (NOS=4-6).

Meta-analysis

Data from 12 studies [4, 11-15, 20, 23, 27, 29] were analysed in a random-effects model to compare the serum/plasma Zn levels in hypothyroid patients and healthy controls. As indicated in Figure 2, pooled effect size revealed that serum Zn levels in subjects with hypothyroidism were not statistically different with those of healthy controls (Hedges' g= -0.84; 95% CI = [-1.74, 0.06]; P = 0.06). However, significant heterogeneity was detected across the studies (I^2=97.5 %, P<0.001). Despite the classification of studies based on disease severity, we could not detect potential source of observed heterogeneity. However, significant differences in pooled effect size of Zn levels were observed in studies conducted on overt hypothyroidism (Hedges' g= −1.19; 95% CI = [−2.33, -0.04]; P = 0.04), while there were non-significant differences in subjects with subclinical hypothyroidism (Hedges' g= 0.15; 95% CI = [-1.23, 1.53]; P = 0.83). The results of subgroup analysis are shown in **Figure 2**. In addition, sensitivity analysis showed that the exclusion of Shah-a study [27] (Hedges' g: -1.02; 95% CI: -1.99, -0.11), Shah-b study [27] (Hedges' g: -1.04; 95% CI: -1.31, -0.06), and Rasic-Milutinovic study [4] (Hedges' g: -1.12; 95% CI: -2.02, -0.21) from the analysis changed the overall effect. There was no evidence of

publication bias among included studies (P=0.21, Begg's test). Nevertheless, results of Egger's test showed a significant publication bias (P=0.03). However, trim-and-fill analyses yielded results similar to the original.

The pooled effect size of 12 studies [4, 12, 13, 19-21, 24, 32, 33] revealed that serum/plasma Se levels in subjects with hypothyroidism was statistically different with those of healthy controls (Hedges' g= −0.45; 95% CI = [−0.88, -0.02]; P = 0.042) **(Figure 3)**. However, significant heterogeneity was detected across the studies (I^2=92.2, P<0.001). Subgroup analysis, based on disease severity, could not detect any potential source of observed heterogeneity. However, significant differences in pooled effect size of Se levels were observed in studies conducted on subclinical hypothyroidism (Hedges' g= −0.51; 95% CI = [-0.86, -0.16]; P = 0.004), while there were non-significant differences in subjects with overt hypothyroidism (Hedges' g= -0.53; 95% CI = [-1.44, 0.37]; P = 0.249) **(Figure 3)**. In addition, sensitivity analysis showed that the exclusion of Wimmer 2014 study [32] (Hedges' g: -0.46; 95% CI: -0.93, 0.01), Federige a 2017 study [21] (Hedges' g: -0.44; 95% CI: -0.90, 0.02), Pedersen a 2013 study [19] (Hedges' g: -0.50; 95% CI: -1.02, 0.02), Faisal Rashid 2010 study [13] (Hedges' g: -0.17; 95% CI: -0.43, 0.10), Eham 2008 study [12] (Hedges' g: -0.46; 95% CI: -0.94, 0.02), Nourbakhsh b 2015 study [24] (Hedges' g: -0.45; 95% CI: -0.91, 0.01), Erdal 2008 study [20] (Hedges' g: -0.37; 95% CI: -0.83, 0.06), and Pedersen b 2013 study [19] (Hedges' g: -0.50; 95% CI: -1.02, 0.01) from the analysis changed the overall effect. There was no evidence of publication bias among included studies (P=0.583, Begg's test & P=0.211, Egger's test).

Fourteen studies [3, 13, 15, 17, 18, 20, 22, 28-30, 34] reported data regarding the serum/plasma Mg levels in hypothyroid patients and healthy controls. As indicated in **Figure 4**, pooled effect size revealed that serum Mg levels in subjects with hypothyroidism was not statistically different

with those of healthy controls (Hedges' g= −0.02; 95% CI = [−0.98, 0.94]; P = 0.970). However, significant heterogeneity was detected across the studies (I^2=98%, P<0.001). Subgroup analysis neither changed the results nor detected any potential source of heterogeneity. Moreover, sensitivity analysis showed that the results were not sensitive to any of the studies. Besides, there was no evidence of publication bias among included studies (P=0.622, Begg's test & P=0.582, Egger's test).

Data from 9 studies [4, 11-14, 20, 23, 29] were analysed in a random-effects model to compare the serum/plasma Cu levels in hypothyroid patients and healthy controls. As indicated in **Figure 5**, pooled effect size revealed that serum Cu levels in subjects with hypothyroidism was not statistically different with those of healthy controls (Hedges' g= −0.09; 95% CI = [−0.40, 0.23]; P = 0.586). However, significant heterogeneity was detected across the studies (I^2=78%, P<0.001). Subgroup analysis yielded no change in the results, nevertheless heterogeneity was attenuated in studies conducted in overt hypothyroidism (I^2=83.3%, P<0.001; Figure 5). Findings from the sensitivity analysis revealed that the exclusion of any single study from the analysis did not alter the overall effect. Besides, there was no evidence of publication bias among included studies (P=0.677, Begg's test & P=0.896, Egger's test).

The pooled effect size from 8 studies [4, 11, 20, 25, 26, 28] showed that serum/plasma Fe levels in subjects with hypothyroidism were not statistically different with those of healthy controls (Hedges' g= −0.77; 95% CI = [−1.55, 0.01]; P = 0.053) **(Figure 6)**. However, significant heterogeneity was detected across the studies (I^2=97.6 %, P<0.001). Despite the classification of studies based on disease severity, we could not detect potential source of observed heterogeneity. However, significant differences in pooled effect size of Fe levels were observed in studies conducted on subclinical hypothyroidism (Hedges' g= −1.11; 95% CI = [-2.21, -0.02]; P =

0.046), while there were non-significant differences in subjects with overt hypothyroidism (Hedges' g= -0.56; 95% CI = [-1.76, 0.64]; P = 0.362) **(Figure 6).** In addition, sensitivity analysis showed that the exclusion of Hanif 2017 [11] (Hedges' g: -1.05; 95% CI: -1.68, -0.43) from the analysis changed the overall effect. There was no evidence of publication bias among included studies (P=0.216, Begg's test & P=0.258, Egger's test).

Four studies [4, 11-13] reported data regarding the serum/plasma Mn levels in hypothyroid patients and healthy controls. As indicated in **Figure 7**, pooled effect size revealed that serum Mn levels in subjects with hypothyroidism was not statistically different with those of healthy controls (Hedges' g= 0.04; 95% CI = [−1.37, 1.44]; P = 0.960). However, significant heterogeneity was detected across the studies (I^2=97.4%, P<0.001). In addition, sensitivity analysis showed that the exclusion of Faisal Rashid (2010) study [13] (Hedges' g: 0.87; 95% CI: 0.03, 1.71) from the analysis changed the overall effect. There was no evidence of publication bias among included studies (P>0.999, Begg's test & P=0.675, Egger's test).

The pooled effect size from 4 studies [4, 11, 29] reported data regarding the serum/plasma Pb levels in hypothyroid patients and healthy controls. The pooled effect size revealed that serum Pb levels in subjects with hypothyroidism was not statistically different with those of healthy controls (Hedges' g= 0.04; 95% CI = [−0.83, 0.92]; P = 0.920) (**Figure 8**). However, significant heterogeneity was detected across the studies (I^2=90.8%, P<0.001). Finding from sensitivity analysis indicated that the exclusion of Joao a (2007) study [29] (Hedges' g: 0.51; 95% CI: 0.04, 0.98) from the analysis changed the overall effect. Moreover, there was no evidence of publication bias among included studies (P=0.174, Begg's test & P=0.201, Egger's test).

Discussion

To the author's knowledge, this is the first systematic review and meta-analysis to have investigated the circulating level of trace elements in patients suffering from hypothyroidism. The results obtained from twenty-five observational studies showed lower Se concentration in patients with hypothyroidism compared to the normal subjects. There was no significant difference in the concentration of Zn, Fe, Cu, Mn, Mg and Pb levels between normal group and patients with hypothyroidism. Subgroup analysis based on disease severity was performed to find-out potential sources of heterogeneity. Subgroup analysis revealed that Zn levels was significantly lower in overt hypothyroidism, while serum/plasma Se and Fe levels were significantly lower in subclinical hypothyroidism compared to healthy participants. In addition, overall estimate of effect size was influence by elimination of several studies; sensitivity analysis showed that results changed to a significant value for Zn, Se, Fe, Mn, Pb. Therefore, findings must be interpreted with caution.

Pooled effect size showed lower Se concentration in patients with hypothyroidism. These findings are in keeping with most recent studies that indicate abnormal metabolism of Se in hypothyroidism [12, 13, 19, 20, 32, 35]. These studies reported that there is an association between Se status and thyroid hormone metabolism, in addition, Se levels were reduced in autoimmune thyroid disorders [12, 32]. The results of previous meta-analyses [5, 36, 37] indicated that Se supplementation might improve Hashimoto's thyroiditis; in essence Se decreased antibody levels and dosage of levothyroxine. Moreover, evidence from animal studies [38, 39] revealed that a Se deficient diet characterized by reduced T_3 and elevated T_4 concentrations. The exact mechanism explaining lower Se concentration in hypothyroidism is

not completely understood, however it may conceivably be attributed to the immunomodulatory and anti-proliferative properties of Se [40]. Most thyroid disorders are self-immune diseases, which are characterized by activated immune system and inflammation [24], thus, the systemic inflammatory setting may have decreased Se levels. n the other hand reduced Se concentration may lead to impairment in the immune function of patients with Hashimoto's thyroiditis [24]. Other possible explanations may be attributed to selenium interference in reducing free radicals production and protecting against oxidative stress-associated disorders [41]. The biological role of selenium in redox control and antioxidative defense is exerted by selenoproteins like the glutathione peroxidases, thioredoxin reductases, and other selenoproteins, and as a result, Se deficiency leads to increased oxidative damage to the thyroid gland [42].

Findings in the current study demonstrated a significantly lower level of serum Zn in overt hypothyroid patients compared with controls. Previous clinical trials [43-45] showed a positive beneficial effect of Zn supplementation on FT_3 levels. Zn deficiency reportedly decreases thyroid hormones, but Zn supplementation adversely affected thyroid function and metabolic activity of these hormones. The possible mechanisms which clarify the possible correlation between Zn deficiency and hypothyroidism might be due to impairment of gastrointestinal absorption of Zn in hypothyroidism patients. Additionally, alteration in Zn distribution was apparent, for instance, increased absorption by other tissues including the liver, which reflects low levels of Zn in these patients [46]. In addition, Zn is necessary for production and activity of thyrotropin releasing hormone (TRH), thus increased TRH concentration in patients with hypothyroidism could be another explanation [7]. Moreover, Zn facilitates T3 binding to the nuclear receptor, indeed, it functions as a coordinating cation for DNA binding proteins [7, 47]. However, the sensitivity

analysis found that after exclusion of Shah study [27] and Rasic-Milutinovic study [4] results changed; therefore, the obtained results should be interpreted cautiously.

We found decreased Fe concentration in subgroup of subclinical hypothyroidism compared to healthy human adults. One possible explanation for these findings could be due to reduced metabolism of Fe in people with hypothyroidism, disturbed gastrointestinal absorption of Fe and subsequently low levels of Fe in hypothyroid patients [48]. On the other hand, thyroid hormones increase production of erythropoietin through direct effect on hematopoiesis [49]. Banday et al. [50] determined that 20-60% of people with hypothyroidism have anemia; as a result, Fe deficiency and reduced thyroid hormones both affect each other's [50]. Sensitivity analysis revealed that results after exclusion of Hanif et al. study [11], changed to a significantly lower value in hypothyroidism patients compared with healthy subjects. Thus interpretation of these results should be drawn with caution.

The present study found no significant difference in serum levels of Mn, Pb, Cu and Mg between two groups; which could be due to high heterogeneity of studies, small sample size of included studies, and differences in populations regarding disease activity. However, it must be noted that exclusion of Faisal Rashid (2010) [13] and Joao a (2007) study [29] changed results to a significantly higher concentration of Mn and Pb in hypothyroid patients compared with controls, respectively. Mn modulates thyroid secreting hormone (TSH) secretion indirectly by dopaminergic pathway [51]. Based on reported evidence the accumulation of Mn in the pituitary gland causes reduced thyroid hormones and elevated TSH concentration [52]. Pb accumulation in the thyroid gland leads to thyroid dysfunction [53], and might act through impairment of the pituitary–thyroid axis and oxido-reductive processes [53]. However more specific studies are needed to explain the exact correlation between these elements and hypothyroidism.

Limitations

The present systematic review and meta-analysis has some limitations that must be acknowledged. Limitations include, the high heterogeneity of included studies regarding study populations and disease severity, the small number of included studies, the small sample size, and other lifestyle and genetic background of participants which could affect final results. We were unable to explore priority or posteriority communication between trace elements and hypothyroidism due to lack of longitudinal studies.

Conclusion

The present meta-analysis found lower Se concentration in patients with hypothyroidism, whereas no significant differences in Cu, Mg, Mn, and Pb were detected between the two groups. In addition, we found decreased concentrations of Zn in overt hypothyroid patients and decreased Fe and Se level in subclinical hypothyroidism. Further studies with higher quality and large sample sizes are required to explicate the link between trace element and hypothyroidism.

Funding

This research was supported by the student research committee of Isfahan university of Medical Sciences [grant number IR.MUI.RESEARCH.REC.1397.433].

Declarations of interest

None

References

[1] N.N. Al-alawae, Diagnosis of Hypothyroidism by Using Some Biomarkers and Immune Markers, Journal of Global Pharma Technology (2018).
[2] P.N. Taylor, D. Albrecht, A. Scholz, G. Gutierrez-Buey, J.H. Lazarus, C.M. Dayan, O.E. Okosieme, Global epidemiology of hyperthyroidism and hypothyroidism, Nature Reviews Endocrinology (2018).
[3] T.Y. Susanna, A. Sagayaraj, K.N. Shashidhar, M. Gomathi, V. Mahesh, A correlative study of thyroid profile and mineral status in patients with hypothyroidism - A hospital-based case control study, 2016.
[4] Z. Rasic-Milutinovic, D. Jovanovic, G. Bogdanovic, J. Trifunovic, J. Mutic, Potential influence of selenium, copper, zinc and cadmium on l-thyroxine substitution in patients with hashimoto thyroiditis and hypothyroidism, Experimental and Clinical Endocrinology & Diabetes 125(02) (2017) 79-85.
[5] K.A. Toulis, A.D. Anastasilakis, T.G. Tzellos, D.G. Goulis, D. Kouvelas, Selenium Supplementation in the Treatment of Hashimoto's Thyroiditis: A Systematic Review and a Meta-analysis, Thyroid 20(10) (2010) 1163-1173.
[6] S.Y. Hess, The impact of common micronutrient deficiencies on iodine and thyroid metabolism: the evidence from human studies, Best Practice & Research Clinical Endocrinology & Metabolism 24(1) (2010) 117-132.
[7] A.E. Pekary, H.C. Lukaski, I. Mena, J.M. Hershman, Processing of TRH precursor peptides in rat brain and pituitary is zinc dependent, Peptides 12(5) (1991) 1025-1032.
[8] J.E. Morley, J. Gordon, J.M. Hershman, Zinc deficiency, chronic starvation, and hypothalamic-pituitary-thyroid function, The American journal of clinical nutrition 33(8) (1980) 1767-1770.
[9] B. Czarnocka, J. Ruf, M. Ferrand, P. Carayon, S. Lissitzky, Purification of the human thyroid peroxidase and its identification as the microsomal antigen involved in autoimmune thyroid diseases, FEBS letters 190(1) (1985) 147-152.
[10] M. Azami, N. Parizad, K. Sayehmiri, Prevalence of Hypothyroidism, Hypoparathyroidism and theFrequency of Regular Chelation Therapy in Patients with Thalassemia Major in Iran: A Systematic Review and Meta-analysis study, Iranian Journal of Pediatric Hematology and Oncology 6(4) (2016) 261-276.
[11] S. Hanif, A. Ilyas, M.H. Shah, Statistical Evaluation of Trace Metals, TSH and T 4 in Blood Serum of Thyroid Disease Patients in Comparison with Controls, Biological trace element research 183(1) (2018) 58-70.
[12] Y.H. Tahs, H.K.A. Hussein, E.A. Ali, Estimation of Serum Copper, Manganese, Selenium, and Zinc inHypothyroid Patients, Journal of the Faculty of Medicine 50(2) (2008) 255-260.
[13] N.F. Rashid, B.A. Abed, T.A. Abas, Relationship between some trace elements, lipid profile and hypothyroidism, Al-Mustansiriyah Journal for Pharmaceutical Sciences 8(2) (2010) 127-138.
[14] S. Baloch, A.R. Memon, A.S. Hayat, N. Masood, Evaluation of serum copper and zinc in hypothyroidism patients, Journal of Science and Technology 3(3) (2013) 316-318.
[15] F. un Nisa, A. Mumtaz, M.I. Ullah, M. Atif, W. Sami, Determination of serum zinc and magnesium levels in patients with hypothyroidism, (2013).

[16] G. Wells, B. Shea, D. O'Connell, J. Peterson, V. Welch, M. Losos, NewCastle–Ottawa quality assessment scale—case control studies, 2017.
[17] A.A. Abdel-Gayoum, Dyslipidemia and serum mineral profiles in patients with thyroid disorders, Saudi Medical Journal 35(12) (2014) 1469.
[18] H.K. Al-Hakeim, Serum levels of lipids, calcium and magnesium in women with hypothyroidism and cardiovascular diseases, Journal of laboratory physicians 1(2) (2009) 49.
[19] I. Bülow Pedersen, N. Knudsen, A. Carlé, L. Schomburg, J. Köhrle, T. Jørgensen, L.B. Rasmussen, L. Ovesen, P. Laurberg, Serum selenium is low in newly diagnosed G raves' disease: a population-based study, Clinical endocrinology 79(4) (2013) 584-590.
[20] M. Erdal, M. Sahin, A. Hasimi, G. Uckaya, M. Kutlu, K. Saglam, Trace element levels in hashimoto thyroiditis patients with subclinical hypothyroidism, Biological trace element research 123(1-3) (2008) 1.
[21] M.A.F. Federige, J.H. Romaldini, A.B.P.P. Miklos, M.K. Koike, K. Takei, E.d.S. Portes, Serum selenium and selenoprotein-P levels in autoimmune thyroid diseases patients in a select center: a transversal study, Archives of endocrinology and metabolism 61(6) (2017) 600-607.
[22] M.G. Gohel, A.M. Shah, J. Makadia, A study of serum calcium, magnesium and phosphorous level in hypothyroidism patients, International Journal of Medical and Health Sciences 3(4) (2014) 308-312.
[23] A. Khubchandani, V. Parmar, V. Solanki, J. Jadav, A. Gamit, S. Gangwani, Comparison of Serum Zinc and Serum Copper in Hypothyroid Patients with Normal Subjects.
[24] M. Nourbakhsh, F. Ahmadpour, B. Chahardoli, Z. Malekpour-Dehkordi, M. Nourbakhsh, S.R. Hosseini-Fard, A. Doustimotlagh, A. Golestani, M. Razzaghy-Azar, Selenium and its relationship with selenoprotein P and glutathione peroxidase in children and adolescents with Hashimoto's thyroiditis and hypothyroidism, Journal of Trace Elements in Medicine and Biology 34 (2016) 10-14.
[25] H. RASHIDIAN, K. SHAHANIPOUR, INVESTIGATING THE SERUM LEVELS OF GLUTATHIONE AND IRON AND THE ACTIVITY OF CASPASE 3 IN PATIENTS WITH HASHIMOTO'S THYROIDITIS, JOURNAL OF ISFAHAN MEDICAL SCHOOL 33(334) (2015).
[26] B. Refaat, Prevalence and characteristics of anemia associated with thyroid disorders in non-pregnant Saudi women during the childbearing age: a cross-sectional study, Biomed J 38(4) (2015) 307-16.
[27] J. Shah, M.R. Jan, M. Yousaf, EFFECTS OF THYROIDAL DYSFUNCTIONS ON BONE MARKERS IN NON-PREGNANT WOMEN: A HOSPITAL BASED CROSS SECTIONAL STUDY FROM KHYBER PAKHTUNKHWA NORTHERN PAKISTAN, Journal of Postgraduate Medical Institute (Peshawar-Pakistan) 31(2) (2017).
[28] S. Shrivastava, S. Shrivastava, AN EVALUATION OF SERUM ELECTROLYTES LEVEL AMONG PATIENTS HAVING HYPOTHYROIDISM, INTERNATIONAL JOURNAL OF SCIENTIFIC RESEARCH 7(5) (2018).
[29] J.B. Souza, J.B. Rocha, C.W. Nogueira, V.C. Borges, R.R. Kaizer, V.M. Morsch, V.L. Dressler, A.F. Martins, É.M. Flores, M.R. Schetinger, Delta-aminolevulinate dehydratase (δ-ALA-D) activity in diabetes and hypothyroidism, Clinical biochemistry 40(5-6) (2007) 321-325.
[30] D. Sridevi, A.A. Dambal, A.S. Challa, S.K. Padaki, A Study of Serum Magnesium, Calcium and Phosphorus in Hypothyroidism, International Journal of Clinical Biochemistry and Research 3(2) (2016) 236-239.

[31] R. Srikrishna, R. Girishbabu, S. Ramesh, Study of Association of Anemia in Sub-clinical and Overt Hypothyroid Patients, RESEARCH JOURNAL OF PHARMACEUTICAL BIOLOGICAL AND CHEMICAL SCIENCES 6(3) (2015) 368-373.
[32] I. Wimmer, T. Hartmann, R. Brustbauer, G. Minear, K. Dam, Selenium levels in patients with autoimmune thyroiditis and controls in lower Austria, Hormone and Metabolic Research 46(10) (2014) 707-709.
[33] P. Zagrodzki, E. Przybylik-Mazurek, Selenium and hormone interactions in female patients with Hashimoto disease and healthy subjects, Endocrine research 35(1) (2010) 24-34.
[34] B.L. Langdahl, A.G.R. Loft, E.F. Eriksen, L. Mosekilde, P. Charles, Bone mass, bone turnover and body composition in former hypothyroid patients receiving replacement therapy, European journal of endocrinology 134(6) (1996) 702-709.
[35] K. Aihara, Y. Nishi, S. Hatano, M. Kihara, K. Yoshimitsu, N. Takeichi, T. Ito, H. Ezaki, T. Usui, Zinc, copper, manganese, and selenium metabolism in thyroid disease, The American journal of clinical nutrition 40(1) (1984) 26-35.
[36] J. Wichman, K.H. Winther, S.J. Bonnema, L. Hegedüs, Selenium supplementation significantly reduces thyroid autoantibody levels in patients with chronic autoimmune thyroiditis: a systematic review and meta-analysis, Thyroid 26(12) (2016) 1681-1692.
[37] E.J. van Zuuren, A.Y. Albusta, Z. Fedorowicz, B. Carter, H. Pijl, Selenium supplementation for Hashimoto's thyroiditis, Cochrane Database of Systematic Reviews (6) (2013).
[38] G.J. Beckett, A. Russell, F. Nicol, P. Sahu, C. Wolf, J. Arthur, Effect of selenium deficiency on hepatic type I 5-iodothyronine deiodinase activity and hepatic thyroid hormone levels in the rat, Biochemical Journal 282(2) (1992) 483-486.
[39] J.-P. Chanoine, M. Safran, A.P. Farwell, P. Tranter, D.M. Ekenbarger, S. Dubord, S. Alex, J.R. Arthur, G.J. Beckett, L.E. Braverman, Selenium deficiency and type II 5'-deiodinase regulation in the euthyroid and hypothyroid rat: evidence of a direct effect of thyroxine, Endocrinology 131(1) (1992) 479-484.
[40] Z. Huang, A.H. Rose, P.R. Hoffmann, The role of selenium in inflammation and immunity: from molecular mechanisms to therapeutic opportunities, Antioxidants & redox signaling 16(7) (2012) 705-743.
[41] C. Benstoem, A. Goetzenich, S. Kraemer, S. Borosch, W. Manzanares, G. Hardy, C. Stoppe, Selenium and its supplementation in cardiovascular disease—what do we know?, Nutrients 7(5) (2015) 3094-3118.
[42] R. Negro, Selenium and thyroid autoimmunity, Biologics: targets & therapy 2(2) (2008) 265.
[43] S. Mahmoodianfard, M. Vafa, F. Golgiri, M. Khoshniat, M. Gohari, Z. Solati, M. Djalali, Effects of zinc and selenium supplementation on thyroid function in overweight and obese hypothyroid female patients: a randomized double-blind controlled trial, Journal of the American College of Nutrition 34(5) (2015) 391-399.
[44] M.A. Aziz, N.Y. Habil, A.K.S. Diab, Effectiveness of Zinc Supplementation in Regulating Serum Hormonal and Inflammatory Status in Hypothyroidism Patients, Medical Journal of Babylon 13(2) (2016) 347-353.
[45] G.A. Kandhro, T.G. Kazi, H.I. Afridi, N. Kazi, J.A. Baig, M.B. Arain, A.Q. Shah, R.A. Sarfraz, M.K. Jamali, N. Syed, Effect of zinc supplementation on the zinc level in serum and urine and their relation to thyroid hormone profile in male and female goitrous patients, Clinical Nutrition 28(2) (2009) 162-168.

[46] K. Yoshida, Y. Kiso, T. Watanabe, K. Kaise, N. Kaise, Y. Itagaki, M. Yamamoto, T. Sakurada, K. Yoshinaga, Erythrocyte zinc in hyperthyroidism: reflection of integrated thyroid hormone levels over the previous few months, Metabolism 39(2) (1990) 182-186.
[47] A. Anselmet, J. Bismuth, J. Torresani, Triiodothyronine nuclear receptor: Role of histones and DNA in hormone binding, Biochimica et Biophysica Acta (BBA)-Gene Structure and Expression 739(3) (1983) 291-300.
[48] K. Dahiya, M. Verma, R. Dhankhar, V.S. Ghalaut, P. Ghalaut, A. Sachdeva, I. Malik, R. Kumar, Thyroid profile and iron metabolism: mutual relationship in hypothyroidism, (2016).
[49] D. Golde, N. Bersch, I. Chopra, M. Cline, Thyroid hormones stimulate erythropoiesis in vitro, British journal of haematology 37(2) (1977) 173-177.
[50] T.H. Banday, S.B. Bhat, S.B. Bhat, S. Bashir, S. Naveed, Incipient iron deficiency in primary hypothyroidism, Thyroid Research and Practice 15(3) (2018) 138.
[51] O. Soldin, M. Aschner, Effects of manganese on thyroid hormone homeostasis: potential links, Neurotoxicology 28(5) (2007) 951-956.
[52] K. Eder, A. Kralik, M. Kirchgessner, The effect of manganese supply on thyroid hormone metabolism in the offspring of manganese-depleted dams, Biological trace element research 55(1-2) (1996) 137.
[53] N.S. Memon, T.G. Kazi, H.I. Afridi, J.A. Baig, S.S. Arain, O.M. Sahito, S. Baloch, M. Waris, Evaluation of calcium and lead interaction, in addition to their impact on thyroid functions in hyper and hypothyroid patients, Environmental Science and Pollution Research 23(1) (2016) 878-886.