ASSESSMENT OF THYROID HORMONES LEVEL IN PREMENOPAUSAL AND POSTMENOPAUSAL FEMALES

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ABSTRACT

Thyroid dysfunction is the common endocrine disorder and females are more affected than males. The aim of this study was to assess the serum thyroid hormone level in premenopausal and postmenopausal females with thyroid dysfunction and observe the effect of thyroid dysfunction on body weight and menstrual regularity. Total 91 female subjects were included in the study. Serum thyroid hormone levels of TSH, FT3 and FT4 were assessed by ELISA technique. It was observed that serum TSH levels were significantly higher in premenopausal and postmenopausal hypothyroid females groups ($P<0.01$) as compared to the control group with normal thyroid function. Demographic characteristics and disease history of the subject were collected. In hyperthyroid premenopausal females serum TSH level was significantly lower than the control group ($P<0.01$). Serum FT3 level was significantly higher in hyperthyroid premenopausal and postmenopausal females as compared to control group ($P<0.01$). In this study, 80% of premenopausal hypothyroid females and 65% of hyperthyroid females complained about menstrual irregularities, this percentage is high as compared to control subjects, which is 20% ($P<0.01$). Inverse negative correlation was observed between TSH, T3, TSH, and T 4, whereas positive correlation was observed between T3 and T 4. Thyroid dysfunction can lead to menstrual irregularities

Key words: Thyroid dysfunction, hypothyroid, hyperthyroid

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INTRODUCTION

Normal reproductive behavior and physiology is dependent on having essentially normal levels of thyroid hormone. (Topper, 1970). Thyroid hormones play an important role in normal reproductive function both through direct effects on the ovaries and indirectly by interacting with sex hormone binding proteins. Thyroid dysfunction can lead to menstrual irregularities and infertility (Poppe & Glinoer, 2003). Diseases of the thyroid gland are among the most abundant disorders worldwide second only to diabetes (Shruti *et al*., 2008). Onset increases with age and it is estimated that 26% of premenopausal and menopausal women are diagnosed with thyroid disease (Hollowell *et al*., 1994). The prevalence and incidence of thyroid disorders is influenced primarily by sex and age. Thyroid disorders are more common in women than men, and in older adults compared with younger age groups (Cappola and Ladenson, 2003). Overt thyroid dysfunction is uncommon in women less than 40 years old and in men <60 years of age (Roos *et al*., 2007). Hypothyroidism is associated with a broad spectrum of reproductive disorders ranging from abnormal sexual development through menstrual irregularities to infertility. The impact of hypothyroidism on the

menstrual cycle has been identified since the 1950s and leads to changes in cycle length and blood flow (Bals-Pratsch *et al*., 1997).

Joshi *et al*. 1993 found 68% of menstrual abnormalities in 22 women with hypothyroidism compared to only 12% in 49 controls. The most common abnormalities observed by hypothyroid women are changes in character of the uterine bleeding and length of the inter-menstrual interval, prolonged and heavy flow is commonly noted. Krassas *et al*. 1999 reported 23.4% hypothyroid female patients had irregular cycles. (Shruti *etel.* 2008). In adult women, hypothyroidism results in changes in cycle length and amount of bleeding and has been reported in association with the ovarian hyper-stimulation syndrome. In an Indian study, 68.2% of hypothyroid women had menstrual abnormalities, compared to 12.2% of healthy controls (Higham, 1992).

Menstrual flow diminishes or ceases in hyperthyroidism despite persistent ovulatory cyclic ovarian function. The mechanism of changes is unknown (Topper, 1994). Krassas *et al.* 1994 reported irregular cycles in 21.5% of the thyrotoxic patients. Hyperthyroidism in women has been linked to reduced fertility. Sowers *et al*. 2003 reported menopausal symptoms, menstrual cycle bleeding characteristics and reproductive hormones and their association with TSH with irregularities of menstrual cycle.

The aim of the present study is to assess the level of thyroid hormones in premenopausal and postmenopausal thyroid dysfunction females and its relationship with body weight and menstrual cycle. Quantitative measurement of thyroid hormones, TSH, FT_3 and FT_4 was done by using enzyme linked immunoenzymometric assay

MATERIAL & METHODS

Experimental design:

In this cross sectional study, serum levels of TSH, FT_4 & FT_3 were measured in premenopausal and postmenopausal female subjects having thyroid dysfunction and compared the values with comparable control parameters. Age matched normal euthyroid subjects were taken as control. The study was carried out from the period of April to August 2008 at Institution of Nuclear Medicine and Oncology Lahore (INMOL), Pakistan for collection of samples from patients with thyroid dysfunction

A structured questionnaire was used for the collection of information from the patients. With the help of questionnaire, information relevant to the study about patient's personal history, duration of disease and use of medication was taken. Question regarding their menstrual history was included which helped to assess the regularity of menstrual cycle during thyroid dysfunction. Women were considered to be menopausal if more than one year had elapsed since their last menstrual period or if they had undergone surgical menopause. Height was measured in inches while weight was taken in kilograms. From height and weight, BMI was calculated.

$$\textbf{BMI = weight/height } (\textbf{m}^2)$$

Range of BMI for normal weight, underweight and overweight is:

- Underweight --- from 15 to 18.4
- Normal --- 18.5 to 22.9
- Overweight --- from 23 to 27.5

A total of 91 female subjects were included in the study. Among 91 subjects, 15 were control group without having any thyroid dysfunction and not taking any kind of medicine

While 76 female subjects were clinically diagnosed patients who had hypothyroidism, hyperthyroidism based upon physicians diagnosis. Among these 76 female subjects, 31 were hypothyroid females and 45 were hyperthyroid females. Further they were divided into premenopausal hypothyroid, premenopausal hyperthyroid, postmenopausal hypothyroid postmenopausal hyperthyroid, and control groups.

Reference range for TSH in normal adult 0.4 – 5.5 µg/L patients with TSH level below this range were considered to be hyperthyroid patients and patients above this range were considered to be hypothyroid patients.

Reference range for FT3 in normal adults 3.5 – 6.47 pg/L patients having FT3 level below this range were considered as hypothyroid subjects and patients with high FT3 levels than the reference range considered as hyperthyroid subjects.

Reference range for FT4 in normal adults 0.8 – 2.7 µg/L patients having FT4 level below this range were considered as hypothyroid subjects and patients with high FT4 levels than the reference range considered as hyperthyroid subjects.

Sample collection:

Blood samples were collected with the help of professional technicians of INMOL, from the inner side of the elbow by using BD syringes of 5 cc. Then samples were brought to Lahore College in vacuette tubes and incubated at 37 ^{0}C for 40 minutes. Then serum was centrifuged at 3000 rpm for 20 minutes and stored at -40 ^{0}C in ependorfs till the time of assay.

Technique used:

ELISA was performed at Lahore College for Women University, by using Bio Rad, CODA Enzyme Immunoassay, for the estimation of serum TSH, FT_4 and FT_3. Kits of Monobind Company were used. For the quantitative determination of TSH, AccuBind ELISA Microwells Product Code 325-300, for FT3 AccuBind ELISA Microwells Product Code 1325-300 and for FT4 AccuBind ELISA Microwells Product Code 1225-300 were used.

RESULTS

A total of 91 females including premenopausal (n=41) post-menopausal (n=35) with thyroid dysfunction and euthyroid females with normal menstrual cycle (Control group= 15) were assessed for T3, T4 and TSH level. Demographic characteristics of the studies population are presented (Table 1). The number of females in premenopausal hypothyroid females group is 15. Mean age value for hypothyroid females is 30.4 ±9.57 years and mean BMI value is 23.4 ± 4.39 kg/m2. Mean values of hormonal parameters are presented in (Table 4). Mean TSH value for this group is 18.8 ±12.86 µg/dl, mean value for FT3 is 1.05 ±0.87 pg/dl and mean value for FT4 is 0.74 ±0.46 µg/dl ($P<0.01$). Menstrual irregularity in this group is 80 % while 20% have regular menstrual cycles

The total number of females in premenopausal hyperthyroid females group is 26. Mean age value for hyperthyroid females is 32.6 ±9.15 years and mean value for BMI is 20.0 ± 2.08 kg/m2. The mean values for hormonal parameters of this group are given in (Table 4). Mean value for TSH is 0.89 ±0.95 µg/dl, for FT3 mean value is 10.79 ±2.36 pg/dl and mean value for FT4 is 2.93 ±1.10 µg/dl ($P<0.01$). Menstrual irregularities in this group are 65% and 35% females have regular menstrual cycles

The number of individuals in postmenopausal hypothyroid females group is 16. For this group the mean age value is 54 ±6.63 years and mean value for BMI is 23.1 ± 1.62 kg/m2. The mean values for hormonal parameters of this are given in (Table 2). Mean TSH value for postmenopausal hypothyroid females is 10.23 ±3.39 µg/dl, for FT3 value is 2.01 ±0.87 pg/dl and mean value for FT4 is 0.48 ±0.38 µg/dl ($P<0.01$).

The number of individuals in postmenopausal hyperthyroid females group is 19. For this group the mean age value is 46 ±7.02 years and mean value for BMI is 27.3 ± 5.34 kg/m2. The mean values for hormonal parameters of this are given in (Table 3). Mean TSH value for postmenopausal hypothyroid females is 0.37 ±0.88 µg/dl, for FT3 value is 11.64 ±2.22 pg/dl and mean value for FT4 is 3.41 ±0.84 µg/dl ($P<0.01$).

The number of individuals in control subjects with normal thyroid function group is 15. The mean values of physical characteristics of this group are given in (Table 1). For this group the mean age value is 23.8 ±3.18 years and mean value for BMI is 20.27 ± 1.76 kg/m2. The mean values for hormonal parameters of this are given in (Table 2). Mean TSH value for is 3.18 ±0.59 µg/dl, for FT3 value is 3.88 ±0.60 pg/dl and mean value for FT4 is 1.83 ±0.58 µg/dl ($P<0.01$). 20% of females have irregular menstrual patterns.

Table 1: Characteristics of the population

Physical characteristics	Hypothyroid	Hyperthyroid	Control
N	31	45	15
Age (mean ±SD)	42.2 ±16.6	39.5 ±9.33	35.8 ±3.81
Height (ft)	5.3	5.2	5.2
Weight (kg)	61.2	52	51
BMI (mean ±SD)	24.32 ±4.21	23.2 ±0.17	20.27 ±0.76
Marital status			
Married	27	38	9
Unmarried	9	7	6
Disease history			
Diagnosed	24	35	--
New cases	7	10	
Menstrual irregularities	80%	65%	3%

Table 2: Comparison of Thyroid l levels in pre and post-menopausal Female (Control Group)

Hormonal Parameters	Premenopause hypothyroid	Postmenopause hypothyroid	Control
n	15	16	15
TSH µg/L **(mean ±SD)**	18.8 ±12.86	10.23 ±3.39	3.18 ±0.59*
FT_3 pg/dl **(mean ±SD)**	1.05 ± 0.87	2.01 ±0.87	3.88 ±0.60*
FT_4 µg/dl **(mean ±SD)**	0.74 ±0.46	0.48 ±0.38	1.83 ±0.58*

**p<0.05*

Table 3: Mean Thyroid Hormonal levels in hypothyroid pre and post-menopausal Female subjects in comparison to healthy controls

Hormonal Parameters	Premenopause hyperthyroid	Postmenopause hyperthyroid	Control
n	26	19	15
TSH µg/dl **(mean ±SD)**	0.89 ±0.95	0.37 ±0.88	3.18 ±0.59*
FT3 pg/dl **(mean ±SD)**	10.79 ±2.36	11.64 ±2.22	3.88 ±0.60*
FT4 µg/dl **(mean ±SD)**	2.93 ±1.01	3.41 ±0.84	1.83 ±0.58*

**p<0.0*

Figure 1: A comparison of Post and pre-menopausal TSH Value in Thyroid Dysfunction and Control Females

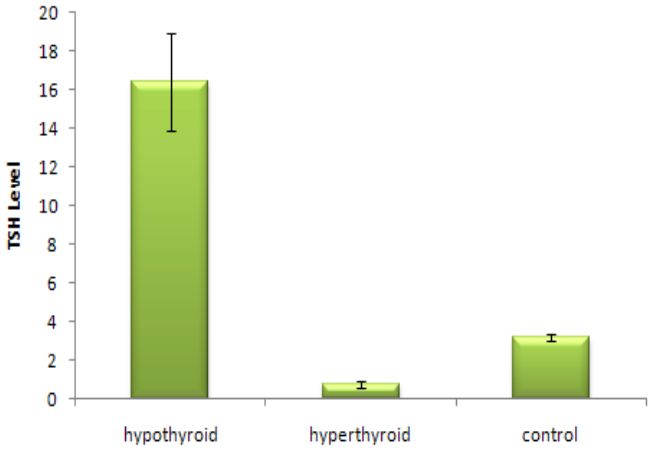


Figure 2: TSH Level in Premenopausal and Postmenopausal Hypothyroid Females

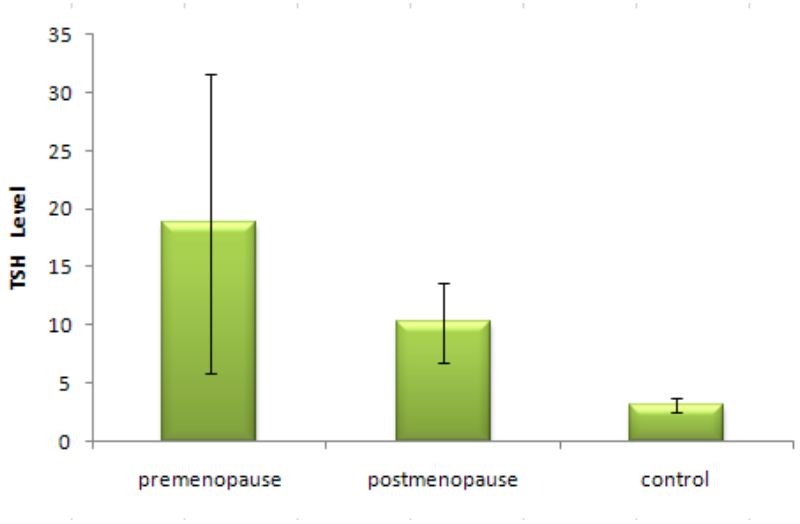


Figure 3: TSH µg/l **in Premenopausal and Postmenopausal Hyperthyroid Females**

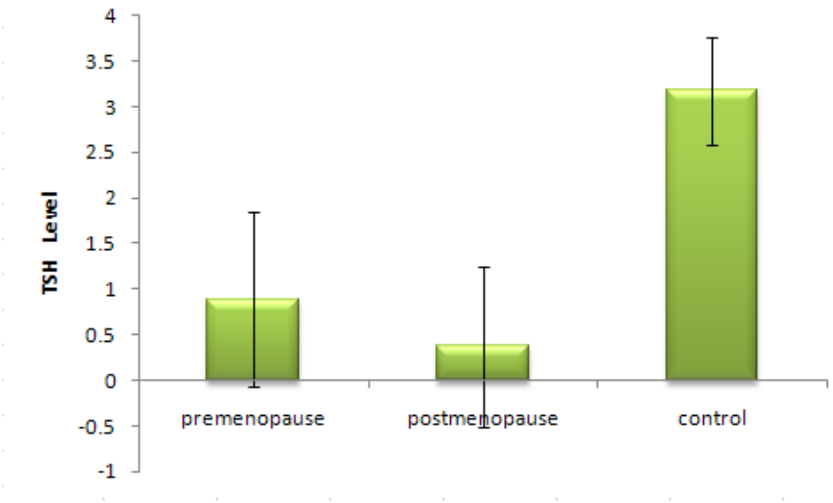


Figure 4: A comparison of fT3 pg/dl in Thyroid Dysfunction and Control Females

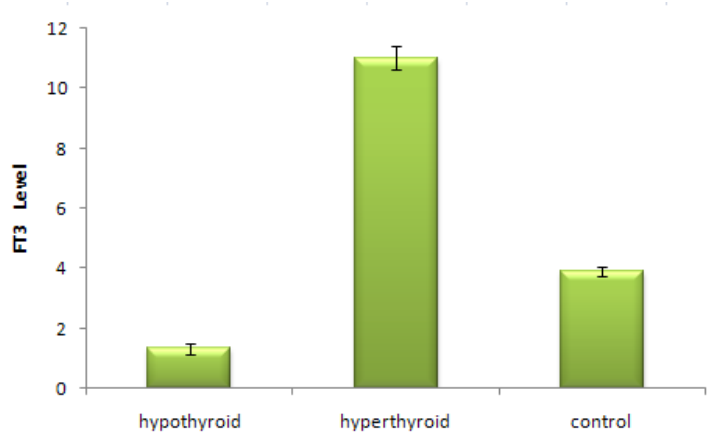


Figure 5: FT3 Level in Premenopausal and Postmenopausal Hypothyroid Females

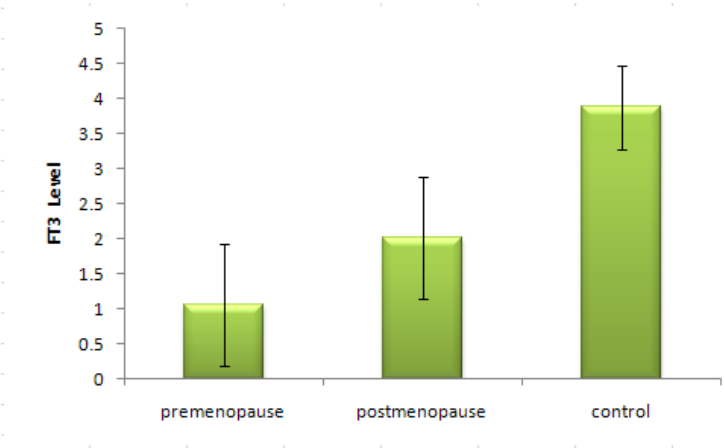


Figure 6: FT3 Level in Premenopausal and Postmenopausal Hyperthyroid Females

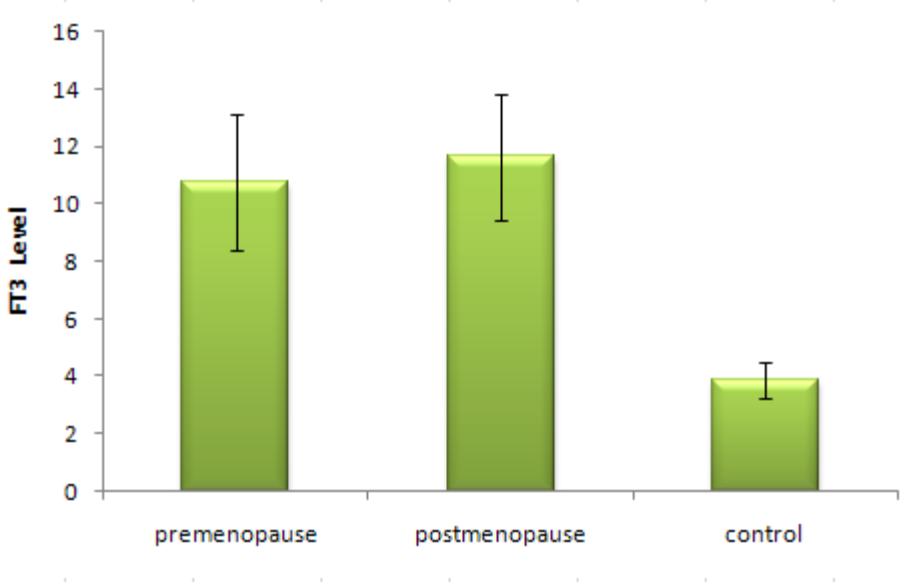


Figure 7: FT4 Level in Thyroid Dysfunction Females

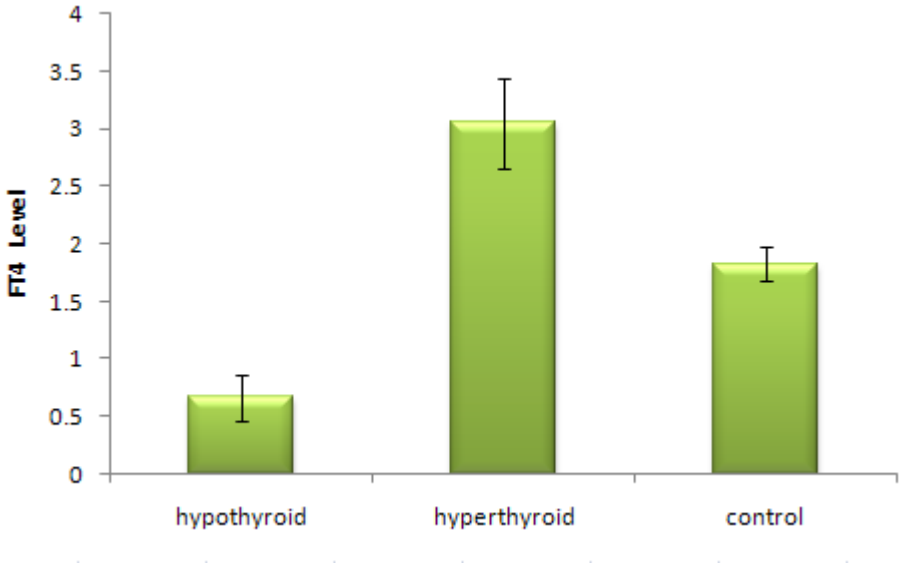


Figure 8: FT4 Level in Premenopausal and Postmenopausal Hypothyroid Females

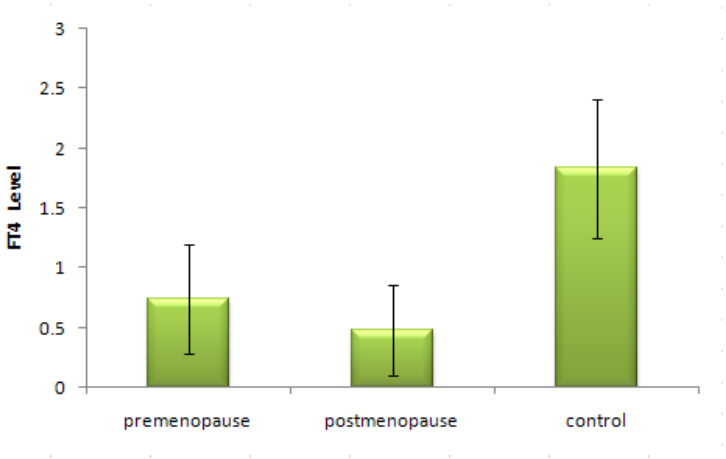
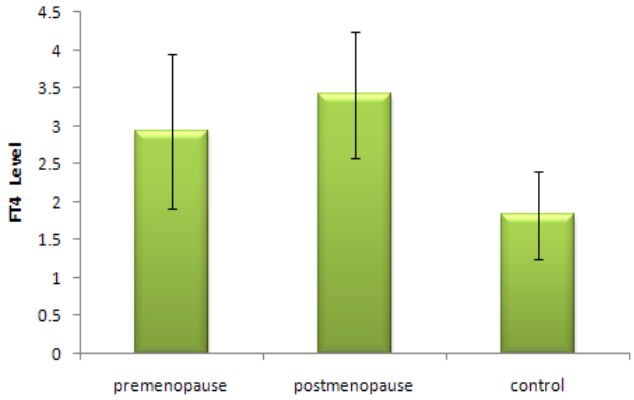


Figure 9: FT4 Level in Premenopausal and Postmenopausal Hyperthyroid Females



DISSCUSION

Diseases of the thyroid gland are among the most abundant disorders worldwide second only to diabetes (Shruti *et al*., 2008). Thyroid disease in general and hypothyroidism in particular are very common in women. Onset increases with age and it is estimated that 26% of premenopausal and menopausal women are diagnosed with thyroid disease (Wartofsky *et al*., 2006). The estimated annual incidence of hyperthyroidism for women ranges from 0.36 to 0.47 per 1,000 women, and for men ranges from 0.087 to 0.101 per 1,000 men. In terms of hypothyroidism, the estimated incidence is 2.4 per 1,000 women each year (Roos *et al*., 2007).

In hypothyroid females gain in body weight was observed having BMI value 24.32 ±4.21kg/m2 ($P<0.01$) which was higher than hyperthyroid and control group. This weight gain in hypothyroidism may be due to the reduction in removal rate of triglycerides and cholesterol which is due to the decrease in the plasma post heparin lipolytic activity. From data obtained general reduction in body weight of hyperthyroid patients was observed. This reduction in weight may be due to the accelerated rate of degradation of most lipids out of proportion to synthesis, so body lipids depots consequently become depleted and levels of various plasma lipid components fall.

Significant difference was observed in TSH levels of premenopausal hypothyroid, hyperthyroid and control group females ($P<0.01$). In premenopausal hypothyroid females elevated TSH level was observed with mean value ($P< 0.05$). Similarly high serum TSH level in postmenopausal hypothyroid females and low serum FT3 and FT4 levels were observed. Increased level of TSH in this group is either due to the decreased secretion of FT3 and FT4 or failure of thyroid gland itself to secrete thyroid hormone. Decreased serum FT3 may be result of decreased conversion of FT4 to FT3.

Similar results were observed by Mortologou and Candilorous, 2004. They reported high serum TSH levels and decreased FT3 and FT4 levels in hypothyroid subjects. Mohanty *et al*. 2008 also reported high levels of TSH in sub-acute hypothyroidism and frank hypothyroidism. Wilson *et al.* 2006, observed low levels of TSH in subclinical hyperthyroid patients While in premenopausal hyperthyroid females decreased level of TSH was ($P<0.01$). In postmenopausal females serum TSH level was low as compared to control group. The increase production of FT3 and FT4 in this group may be due to autoimmune thyroiditis. This increase in thyroid hormone is the one cause of decreased TSH level in hyperthyroidism. Similar results were observed by number of studies. Mortologou and Candilorous, 2004 reported, suppressed serum TSH level and high serum FT3 and FT4 levels in hyperthyroid subjects. Wilson et al. 2006 also reported higher serum FT4 and FT3 levels in hyperthyroid females. Similar decrease in TSH secretion was reported by Lonn et al. 1998 in hyperthyroid subjects, suppressed serum TSH and elevated serum FT3 and FT4 levels were also studied by Mcdermott and Ridgway, 1998 in hyperthyroid subjects.

Normal reproductive behavior and physiology is dependent on having essentially normal levels of thyroid hormone. Hypothyroidism in particular is commonly associated with infertility (Topper, 1994). Thyroid hormones play an important role in normal reproductive function both through direct effects on the ovaries and indirectly by interacting with sex hormone binding proteins. (Poppe & Glinoer, 2003).

In the present study, menstrual abnormalities were observed in premenopausal hypothyroid and hyperthyroid females. Menstrual abnormalities percentage is higher in premenopausal hypothyroid (females 80%) as compared to premenopausal hyperthyroid females (65%). These results are in line with Joshi *et al.*, 1993 and abnormal menstrual pattern was observed in his studies. Krassas et al. 1999 reported (23.4%) hypothyroid female patients had irregular cycles. In this study correlation between the thyroid hormones was also observed. In premenopausal and postmenopausal hypothyroid females non-significant inverse correlation was observed between TSH and fT3 and significant correlation between TSH and fT4 was observed, whereas positive non-significant relation was observed between fT4 and fT3 in premenopausal hypothyroid females. A significant positive correlation was observed between T4 and T3 in postmenopausal hypothyroid females. In premenopausal and postmenopausal hyperthyroid females non-significant inverse correlation was observed between TSH and fT3. Thyroid dysfunction can lead to menstrual irregularities

CONCLUSION

Thyroid dysfunction can lead to menstrual irregularities. It was concluded from the present study that serum TSH levels were significantly higher in premenopausal and postmenopausal hypothyroid females ($p < 0.01$)

REFERENCES

1. Topper, Y. J. 1970. Multiple hormone interactions in the development of mammary gland in vitro. *Recent Progress in Hormone Research*, 26:287
2. Poppe, K. & Glinoer, D. 2003. Thyroid autoimmunity and hypothyroidism before and during pregnancy. *Human Reproduction Update*, 9: 149–161.
3. Shruti, M., Amruthlal, W., Reddy, G. C., Kusumanjali, G., Kanagasabapathy, A. S. and Pragna, R. 2008. Diagnostic strategies for subclinical hypothyroidism. *Indian Journal of Clinical Biochemistry*, 23 (3): 279-282.
4. Hollowell, J. G., Staehling, N. W., Flanders, W. D., Hannon, W. H., Gunter, E. W., Spencer, C. A. and Braverman, L. E. 2002. Serum TSH, T4, and thyroid antibodies in the United States population (1988 to 1994): National Health and Nutrition Examination Survey (NHANES III). *The Journal of Clinical Endocrinology and Metabolism*, 87(2):489–499.

5. Cappola, A. R. and Ladenson, P. W. 2003. Hypothyroidism and Atherosclerosis. *The Journal of Clinical Endocrinology and Metabolism*, 88: 2438-2444.
6. Roos, A., Bakker, S. J., Links, T. P., Gans, R. O. and Wolffenbuttel, B. H. 2007. Thyroid Function is Associated with Components of the Metabolic Syndrome in Euthyroid Subjects. *The Journal of Clinical Endocrinology and Metabolism,* 2: 491-496.
7. Bals-Pratsch, M., Geyter, D., Muller, C., Frieling, T., Lerchl, U., Pirke, A., Hanker, K. M., Becker, J. P., Carus, C. & Nieschlag, E. 1997. Episodic variations of prolactin, thyroid-stimulating hormone, luteinizing hormone, melatonin and cortisol in infertile women with subclinical hypothyroidism. *Human Reproduction*, 12:896.
8. Higham, J. M. 1992. The effect of thyroxine replacement on menstrual blood loss in a hypothyroid patient. *British Journal of Obstetrics and Gynecology*, 99: 695–696.
9. Joshi, J. V., Bhandarkar, S. D., Chadha, M., Balaiah, D. & Shah, R. 1993. Menstrual irregularities and lactation failure may precede thyroid dysfunction or goitre. *Journal of Postgraduate Medicine*, 39:137–141.
10. Krassas, G. E., Pontikides, N. and Kaltsas, T. 1999. Disturbances of menstruation in hypothyroidism. *Clinical Endocrinology*, 50:655-659.
11. Krassas, G. E., Pontikides, N. and Kaltsas, T. 1994. Menstrual disturbances in thyrotoxicosis. *Clinical Endocrinology*, 40:641-644.
12. Sowers, M. F., Luborsky, J., Perdue, C., Katy, L. B., Araujo, M., Goldman, B. and Siobán, D. 2003. Thyroid stimulating hormone (TSH) concentrations and menopausal status in women at the mid-life. *Clinical Endocrinology*, 58(3):340 – 347.
13. Surks, M. I., Ortiz, E., Daniels, G. H., Sawin, C. T., Cobin, R. H., Franklyn, J. A., Burman, K. D., Denke, M. A., Cooper, R. S. and Weissman, N. J. 2004. Subclinical thyroid disease. Scientific review and guidelines for diagnosis and management. *Journal of the American Medical Association*, 291:228–238.
14. Wartofsky, L., Nostrand, D. V. and Burman, K. D. 2006. Overt and subclinical hypothyroidism in women. *Surgery Obstetrics and Gynecology,* 61(8):535-542.
15. Roos, A., Bakker, S. J., Links, T. P., Gans, R. O. and Wolffenbuttel, B. H. 2007. Thyroid Function is Associated with Components of the Metabolic Syndrome in Euthyroid Subjects. *The Journal of Clinical Endocrinology and Metabolism,* 2: 491-496.
16. Bemben, D. A., Winn, P., Hamm, R. M., Morgan, L., Davis, A. and Barton, E. 1994. Thyroid disease in the elderly. Part 1. Prevalence of undiagnosed hypothyroidism. *Journal of Family Practice*, 38(6):577–582.
17. Marqusee, E., Haden, S. T. and Utiger, R. D. 1998. Subclinical thyrotoxicosis. *Endocrinology and Metabolism Clinics of North America*, 27(1):37–49.
18. Lonn, L., Stenlof, K., Ottosson, M., Lindroos, A., Nystrom, E. and Sjostrom, L. 1998. Body Weight and Body Composition Changes After Treatment Of Hyperthyroidism. *The Journal of Clinical Endocrinology and Metabolism,* 83 (12):4269-4273

19. Bayley, T. A., Harrison, J. E., Mcneill, K. G. and Mernagh, J. R. 1980. Effect of Thyrotoxicosis and Its Treatment on Bone Mineral and Muscle Mass. *The Journal of Clinical Endocrinology and Metabolism*, 50:916-922.
20. Poppe, K. & Glinoer, D. 2003. Thyroid autoimmunity and hypothyroidism before and during pregnancy. *Human Reproduction Update*, 9: 149–161.
21. Kumar, M. S., Safa, A. M., Deodhar, S. D. and Schumacher, O. P. 1977. The relationship of thyroid-stimulating hormone (TSH), thyroxine (T4), and triiodothyronine (T3) in primary thyroid failure. *American Journal of Clinical Pathology*, 68(6):747-51.
22. Mcdermott, M. T. And Ridgeway, E. C. 1998. Central hyperthyroidism. *Endocrinology and Metabolism Clinics of North America*, 27(1):187-203.
23. Wilson, S., Parle, V. J., Roberts, M. L., Roalfe, K. A., Hobbs, R. D., Clark, P. and Franklyn, J. A. 2006. Prevalence of subclinical thyroid dysfunction and its relation to socioeconomic deprivation in the elderly: a community based cross-sectional survey. *Journal of Clinical Endocrinology & Metabolism*, 91(12): 4809-4816.