

RESEARCH ARTICLE

Old-Aged, Adolescent and Pregnant Women in Eastern Nepal are more prone to Sub-Clinical Hypothyroidism: A Hospital-Based Retrospective Study**Ram L. Mallick^{1*}, Shripad J. Walavalkar², Subarna Pokhrel¹, Randhir Kumar Singh³,**

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ABSTRACT

Mild thyroid failure or sub-clinical hypothyroidism is a common problem without known thyroid disease. The disorder is more prevalent in women and generality increases with age. In women at reproductive age, sub-clinical hypothyroidism is more common among infertile subjects compared to general population. Miscarriage rates are higher in females having sub-clinical hypothyroidism with TSH levels > 4 mIU/L. Diagnosis of sub-clinical hypothyroidism is thus necessary in these subjects. In eastern part of Nepal, the prevalence of sub-clinical hypothyroidism is approximately 20 %. However, in spite of the predominance, generality of sub-clinical hypothyroidism among adult women was still unknown at this place. Thus we investigated sub-clinical hypothyroidism prevalence in old-aged, adolescent and pregnant women at the same location. Our data demonstrate that aforesaid females exhibit 10.7 % generality. Further 79 % subordinates among affected individuals were under reproductive age (< 45 years). Hence our study reflects the concern to care thyroid problems in this locality.

Key words: Thyroid disease, reproductive age, sub-clinical hypothyroidism.

INTRODUCTION

Hypothyroidism is common disorder that occurs due to deficiency in secretion of thyroid hormone and its action ^[1]. It takes place in mild or severe forms in 2-15 % in females ^[2]. Hypothyroidism can occur in infants and children although its generality is rare in these subjects. If clinicians treat hypothyroidism within first month, a baby grows with proper mental development ^[3]. Untreated hypothyroidism can damage brain and leads to intellectual disability as well as developmental delays. By contrast, intellectual disability fails to appear if hypothyroidism develops after 3-years of age. In USA, clinicians therefore test all children for hypothyroidism at birth. Further, as age advances, undiagnosed hypothyroidism delays physical growth and sexual development including onset of puberty. Also called Hashimoto's thyroiditis is an autoimmune disease in which individual's immune system turns against body's own tissues ^[4]. Sometimes hypothyroidism caused by Hashimoto's thyroiditis

disappear on its own. But more often, the disorder causes gradual loss of thyroid function. The symptoms germinate slowly which is difficult to notice for affected individuals for years. Myxedema is another thyroid-related clinical condition that causes swelling of tissues with fluid retention around heart and lungs ^[5].

Hypothyroidism afflicts more in adult women. There exist 2-4 % prevalence of hypothyroidism in females during reproductive life (< 45-years of age) ^[6,7]. In laboratory medicine, patients suffering from hypothyroidism demonstrate hyponatremia, normocytic anemia, elevated creatine kinase, hyperprolactinemia and hyperlipidemia ^[8]. Clinical symptoms of hypothyroidism range from easy to recognize features such as lethargy, fatigue and cold intolerance to subtler disease (like sub-clinical hypothyroidism (SCH)) with general symptoms that escape detection. SCH is clinically asymptomatic. Patients suffering from SCH do not exhibit symptoms and health difficulty. Certain people having SCH regain

normal thyroid function. But about 10 % people who have SCH can go on to get hypothyroidism within three years^[9]. Thus diagnosis of SCH itself is important. For identification point of view, persons affected from SCH demonstrate slight abnormal thyroid test results. Based on laboratory reports, serum levels of thyroxine (T4) and triiodothyronine (T3) lie within reference range and that of thyroid stimulating hormone (TSH) elevates. Clinicians divide SCH into persons with TSH levels above upper reference limit and those with values between 4 and 4.5 mIU/L. In women at reproductive age, SCH is more common among infertile subjects compared to general population. Miscarriage rates are higher in women having SCH with TSH levels > 4 mIU/L. Elevated serum TSH levels, therefore, correlate to pathologies and parametric study in clinical laboratory is only a diagnostic tool for SCH. Further patients with SCH have high rate of progression to clinically overt hypothyroidism^[10].

Mild thyroid failure or SCH is common problem with prevalence of 3-8 % in population without known thyroid disease^[11]. SCH is more frequent in women. Its prevalence increases with age and is higher in females. In earlier study, Rohil *et al.* reported that SCH is prevalent in approximately 20 % Nepalese people in eastern region of Nepal^[12]. SCH is, therefore, predominant in persons at this part of the world. However, in spite of its predominant generality, SCH prevalence among adult women in same area was unknown. Thus we investigated SCH prevalence in old-aged, adolescent and pregnant women in eastern Nepal. In present study we observed 10.7 % generality of SCH that had surpassed its 3-8 % prevalence in general population. Among these affected subordinates, 79 % were within reproductive-age-limit of 45-years. We, therefore, inferred that women at reproductive age in eastern Nepal are more prone to have SCH. Further our study reflects the concern to care thyroid problems in locality.

MATERIALS AND METHODS

Study design and enrolment criteria:

This was hospital-based retrospective study conducted in the Department of Diagnostic Laboratory at Birat Medical College and Teaching Hospital (BMCTH), Biratnagar, Morang, Nepal. Old-aged, adolescent and pregnant women who attended BMCTH from rural area of eastern Nepal for investigation of SCH were enrolled as subjects in the study. Duplication of a similar participant

and patients on thyroid medication, chronic infection, injury, malignancy and recent history of surgery were excluded. Enrolment of 440 subjects was planned to form target population.

Sample collection and serum preparation:

Venipuncture was performed for collection of blood samples under universal attentiveness as described in manufacturer's protocol. Antecubital venous blood from patients was collected in plain vial with informed written consent, strictly as per the recommendations and approval of the Institutional Ethical Committee. Blood samples were allowed to clot for 5 min and centrifuged at 3000 r.p.m. for 15 min in order to separation of serum. Sera were stored at -20 °C until assessment of thyroid hormones. All steps were carried out under sterile conditions and precautions were taken to prevent blood from hemolysis and free of fibrin.

Estimation of free triiodothyronine (FT3):

FT3 immunoassay was performed using direct chemiluminescent technology (ADVIA Centaur FT3 assay kit) according to instruction of the manufacturers. ADVIA Centaur XP System was employed while performing assay. System was prepared after loading reagent packs (containing monoclonal mouse anti-FT3 antibody). Instrumentation had automatically performed actions that include: dispensing 50 µl serum into cuvette, adding 100 µl Lite reagent (containing 0.1 % sodium azide) followed by incubation at 37 °C for 5 min, supplementing 450 µl Solid Phase reagent and incubating at 37 °C for 2 min 30 s. System had separated, aspirated and washed the cuvettes with reagent water. Following incubation, system had dispensed 300 µl each of acid and base reagents to initiate chemiluminescent reaction. Results were obtained according to selection option as stated in system operating instructions. Based on setting up of the assay, system had reported FT3 results in pg/ml.

Estimation of free tetraiodothyronine (FT4):

FT4 immunoassay was performed using direct chemiluminescent technology by FT4 assay kit (from manufacturer as mentioned under estimation of FT3). With application of an analyser (that was employed above for estimation of FT3) and after system was loaded with reagent packs (containing monoclonal mouse anti-FT4 antibody), estimation based on chemiluminescent reaction was carried out as discussed. System was set to receive FT4 results in ng/dl.

Estimation of TSH:

TSH immunoassay was performed using direct chemiluminescent technology by TSH assay kit as mentioned above and according to instructions of manufacturers. After loading an Analyzer with reagent packs (containing monoclonal mouse anti-TSH antibody), experiment was performed. System dispensed itself 200 µl serum in cuvette, added 50 µl Lite reagent and 225 µl Solid Phase followed by incubation at 37 °C for 7 min 30 s. Separation, aspiration, washing of cuvettes and chemiluminescent reaction steps were in accordance with FT3 and FT4 assays. Assessment was fixed to acquire TSH results in mIU/L.

Data interpretation:

The validity and reliability of test results were determined using control sera (from *BIO-RAD*). Data were analyzed under Software Package for Social Sciences version 16 (SPSS 16).

RESULTS

SCH prevalence was significant among total patients:

SCH is prevalent in population without known thyroid disease. We, therefore, collected antecubital venous blood and separated sera to determine thyroid status among patients. As higher serum TSH with upper reference limit despite of normal levels of thyroxine characterize SCH, we performed FT3, FT4 and TSH immunoassay. As expected, control sera confirmed 2.8 ± 1.4 pg/ml, 1.4 ± 0.6 ng/dl and 3.25 ± 2.85 mIU/ml as reference ranges for FT3, FT4 and TSH, respectively (data not shown). Out of total patients ($n=440$), major portion ($n=338$) demonstrated FT3, FT4 and TSH within reference ranges, suggestive of normal functioning and secretion of thyroid gland i.e., euthyroidism in 76.8 % population (figure 1A; fade-coloured bar).

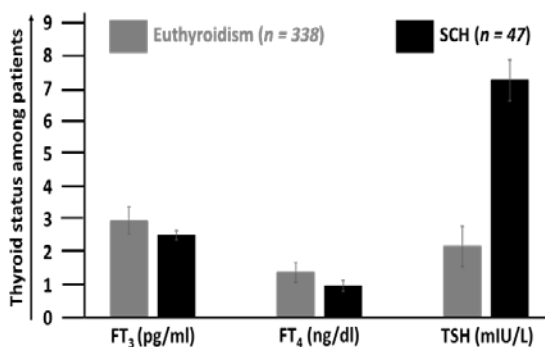


Figure 1 A. Bar diagram representing circulating levels of FT₃, FT₄ and TSH among attending subjects. Symbols within brackets indicate units of measurement of corresponding parameters.

remains to be established. However, due to high prevalence of SCH and associated metabolic risk factors such as hyperlipidemia, the American Thyroid Association recommends to begin measurement of serum TSH for screening purpose at age 35 years and every 5 years thereafter. We adapted as case of SCH when patients showed TSH levels more than 6.1 mIU/ml. Out of $n=440$, 47 patients demonstrated TSH levels > 6.1 mIU/ml, indicating that significantly 10.7 % suffered from SCH (figure 1A; black-coloured bar).

Remaining 55 victims (12.5 %) with TSH levels > 50 mIU/ml validated hypothyroidism (figure 1B).

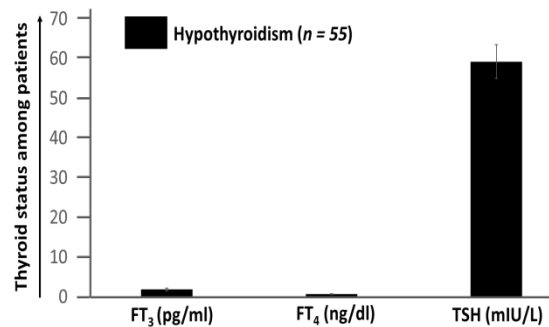


Figure 1 B. Bar diagram representing circulating levels of FT₃, FT₄ and TSH among attending subjects. Symbols within brackets indicate units of measurement of corresponding parameters.

SCH was predominant in late adults:

We observed 10.7 % people suffered from SCH (figure 1A; black-coloured bar).

By contrast, our data showed the persons having SCH were in age groups between 15- and 65-years-old. We, therefore, separated subjects with SCH into five sets to analyze prevalence against age variation. Out of 47 patients having SCH, we observed 4 and 3 were within 15-25 and 56-65 years-old-age, respectively (figure 3; 1st and 5th bars).

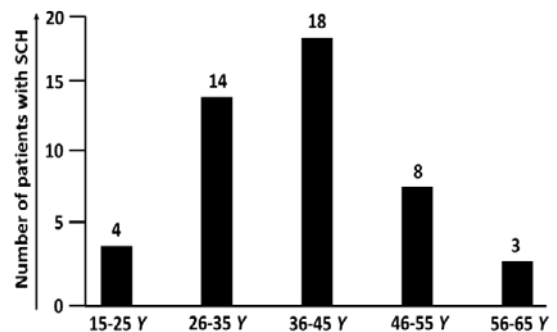


Figure 3. Bar diagram representing number of patients suffering from SCH as age wise (Y = years-age group)

Uniform national guidelines for screening of thyroid disease with serum TSH levels still

Prevalence of SCH among 15-25 and 56-65 year-old patients thus remained less than 10 % in separate, indicative of minor existence of SCH among teenager adults and aged people. Further

14 and 8 individuals of ages 26-35 and 46-55 years, respectively, ensured SCH (figure 3; 2nd and 4th). As 26-35 and 46-55 year-old patients grasped, respectively, about 30 and 17 % fraction, our data specified that disease prevalence is significant in adult SCH population. Interestingly, when we had look upon a residual group holding patients of ages 36-45 years, 18 (out of 47) i.e., immensely 38 % were sufferers (figure 3; 3rd bar), suggestive of predominant existence of SCH in late adults.

SCH was 10 and 12 % incidental in adult- and elderly-women, respectively:

SCH is common problem. Prevalence of SCH increases with age and is higher in women. Thus, in order to inquiry of the SCH incidence among adult- and elderly-individuals, we separated females into two groups. Attending women below 45-years were considered as adults and similar counterparts above mentioned-age as elderly people. Out of 440 patients, approximately 79 % (n=347) stood below and 21 % (n=93) persisted above 45-years (figure 4), suggesting that mostly adult women had complaints of thyroid disorders.

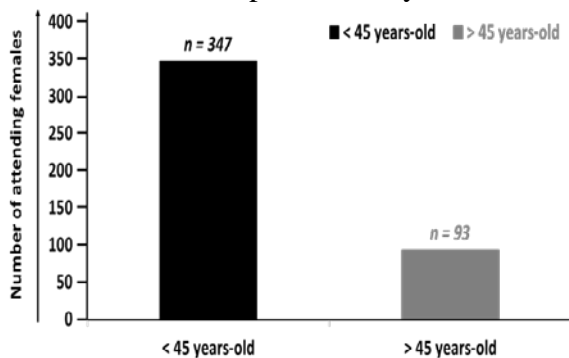


Figure 4. Bar diagram to compare number of attended subjects as stage-wise (adult and aged)

Next, we categorized our findings to specify the marks. Among 347 subjects underlying 45-years of age (figure 4; black-coloured bar); functioning and secretion of thyroid gland appeared normal in 78 % (n=270) people (figure 5; black-coloured bar in 1st set),

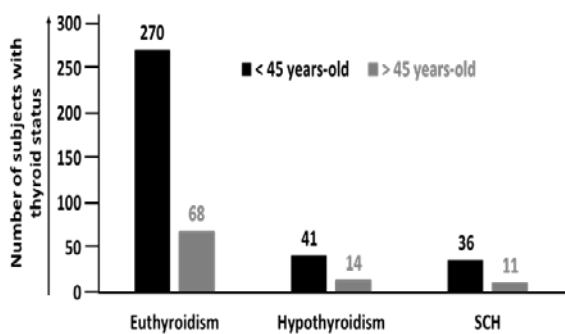


Figure 5. Bar chart to compare thyroid status across stage-wise (adult and aged)

around 12 % (n=41) (figure 5; black-coloured bar in 2nd group) suffered from hypothyroidism and remaining 10 % (n=36) (figure 5; black-coloured bar in 3rd unit) had mild thyroid failure or SCH. Further among 93 elderly-women who crossed 45-years (figure 4; shaded-bar), thyroid status was usual in approximately 73 % (n=68) (figure 5; shaded-bar in 1st set), another 15 % (n=14) (figure 5; shaded-bar in 2nd group) underwent hypothyroidism and residual 12 % (n=11) (figure 5; shaded-bar in 3rd unit) fit in SCH, thus indicative of about 10 % incidence of SCH in adult- and 12 % among elderly-women.

DISCUSSION

In present study we demonstrate that SCH is prevalent in approximately 10.7 % old-aged, adolescent and pregnant women in eastern Nepal (figures 1A and 2). Based on certain previous studies conducted in Nepal, SCH was prevalent in more than 20 % Nepalese [12, 13]. By contrast, in eastern part of the nation, frequency of SCH among old-aged, adolescent and pregnant women was unknown. Thus we surveyed SCH prevalence in aforementioned females at same region and observed 10.7 % generality (figures 2). SCH or mild hypothyroidism occurs in 3-8 % in general population. In USA, according to National Health and Examination Survey-III (NHANES-III) that excluded subjects with known thyroid diseases, approximately 4.5 % Americans suffered from SCH. The data about 10.7 % prevalence in present study had surpassed 3-8 % generality in normal people. Our findings, therefore, recommend for the information that eastern Nepalese females are more prone to have SCH. Further, 10.7 % prevalence reflects to care about frequent thyroid problems in locality.

Iodine deficiency and overt hypothyroidism during pregnancy contribute to adverse fetal neurologic effects. Screening for and treating SCH during pregnancy is, therefore, necessary. However, to screen for and treat SCH during pregnancy has been a theme of ongoing debate and research in last decade. In the present study, we observed 79 % prevalence of SCH (figure 4; black-coloured bar) in women below 45-years and that in females above mentioned-age was 21 % (figure 4; shaded-bar). Moreover, among 47 patients who suffered from SCH, the age of 14 and 18 individuals were 26-35 and 36-45 years. Thus a significant total of 30 and 38 % Nepalese females were sufferers at particularly during reproductive-age. Our data regarding more

prevalence of SCH in women below 45-years in our study, it is plausible that females who were below 45-years and having SCH underwent successful treatment. Still, accurate explanation behind more prevalence of SCH in women below 45-years remains an area of research in same orbit. Several pieces of studies supported that hypothyroidism has a link to depression^[15-18]. In future, analytical approach to investigate origin of SCH prevalence is therefore needful and will grip the thyroid concern among old-aged, adolescent and pregnant women in eastern part of Nepal.

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