Original Article

Association of Prehypertension Status with Cardiovascular Risks in Subclinical Hypothyroidism

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Abstract

Background and Aim: Although attenuated baroreflex sensitivity (BRS) is known to promote cardiovascular (CV) risk, its status in subclinical hypothyroidism (SCH) has not been reported. Furthermore, the link of sympathovagal imbalance (SVI) to CV risk in SCH has not been reported. This study aimed to assess the association of BRS to hypertension status and CV risk in SCH. **Methods:** SCH patients (n = 32) and controls (n = 32) were recruited for the study. Body mass index, basal heart rate (HR), systolic blood pressure (BP), diastolic BP, BRS, autonomic function tests, HR variability (HRV), high-sensitive C-reactive protein, lipid profile, and atherogenic index were measured. Prediction of prehypertension status by BRS was assessed. **Results:** There was significant decrease in BRS, increased sympathetic and decreased parasympathetic reactivity, decreased HRV and high-density lipoprotein, and significant increase in all other lipid parameters, in SCH compared to controls. A significant association was found between BRS and low-frequency to high-frequency ratio in SCH group. BRS had a significant prediction of prehypertension status. **Conclusion:** SVI and decreased BRS were observed in SCH group that was linked to SVI. Reduced BRS that contributes to prehypertension status predisposes to subclinical hypothyroids to CV risks.

Keywords: Autonomic functions, baroreflex sensitivity, cardiovascular risks, prehypertension status, subclinical hypothyroidism, sympathovagal imbalance

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Introduction

Among endocrine disorders, hypothyroidism is second to diabetes mellitus in prevalence. In India, prevalence of overt hypothyroidism (OH) is 10.95% while the prevalence of subclinical hypothyroidism (SCH), an unquantified cause of morbidity, is reported to be between 3% and 15%.[1,2] The presentation of OH varies from vague symptoms such as tiredness and weight gain to pericardial effusion and cardiac dysfunction, indicating a wide spectrum in terms of presentation and severity. The cardiovascular (CV) dysfunction in OH is well documented and is explained on the basis of decreased action of thyroid hormones on the CV system. The impact of SCH as an entity on CV morbidity of a community is underestimated; hence, numerous studies have been done in this regard. It is documented that patients with SCH invariably progress to OH and are also at CV risk, supported by echocardiograph evidence.[3] Autoimmune thyroiditis, which is a major cause of thyroid dysfunction, is associated with low-grade inflammation that predisposes to atherosclerosis.[4] Profound hyperlipidemia is an established biochemical phenomenon in hypothyroidism. Dyslipidemia in OH causes oxidative stress, which directly contributes to CV morbidity.[5]

Hypothyroidism is considered to be a hypometabolic state. A report indicated reduced sympathetic drive and an increase in vagal activity.[6] However, studies from our laboratory have revealed increased sympathetic drive in both hypothyroidism and hyperthyroidism contradicting earlier findings, which contributes to sympathovagal imbalance (SVI), with an increased sympathetic activity coupled with reduced vagal activity.[7,8] Further, contribution of SVI to CV risk in SCH has

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not yet been documented. Conventional autonomic function tests (CAFTs), heart rate variability (HRV), and blood pressure variability (BPV) analysis have been used as noninvasive tools objectively to detect CV risks and to diagnose autonomic dysfunction at an early stage.[9,10] Therefore, this study was designed to assess the role of inflammation and dyslipidemia as causal factors for CV risks and correlate it with SVI in SCH and also to assess the association of prehypertension status with baroreflex sensitivity (BRS), the marker of CV risk, in SCH.

Materials and Methods

This was an analytical case–control study conducted in subclinical hypothyroid group, designed to assess the CV function and autonomic status in subclinical hypothyroid group in comparison to age- and gender-matched healthy individuals and to assess the magnitude of cardiac dysfunction in relation to their biochemical profile and CV autonomic status. The study was conducted in the Autonomic Function Testing Laboratory, Department of Physiology, JIPMER. The sample size was estimated at 5% level of significance and 80% power using OpenEpi software (Version 2.3.1), in which the sample size was calculated to be 32. The approval of the Institute Research Council and Institute Ethics Committee for Human Studies was obtained before the commencement of the study.

Recruitment and grouping of subjects

Study group (cases)

Thirty-two cases were included in the study.

The cases were recruited from the outpatient unit of the Endocrinology Department of JIPMER.

- Inclusion criteria
  - Freshly diagnosed cases of hypothyroidism aged between 18 and 45 years before initiation of treatment were included
- Exclusion criteria
  - Hypothyroid patients receiving thyroxin replacement therapy
  - Known cases of diabetes mellitus, hypertension, heart disease, autonomic failure, and any other endocrine disorders or on any hormonal therapy or drugs affecting autonomic function.

Control group

Thirty-two individuals were recruited.

- Inclusion criteria: Healthy age- and gender-matched normal individuals
- Exclusion criteria: Individuals suffering from any illness such as diabetes mellitus, hypertension, heart disease, autonomic failure, and any other endocrine disorders or on any hormonal therapy or drugs affecting autonomic function.

Written informed consent was obtained from all the participants before the commencement of the study.

Study procedure

The individuals were asked to report to autonomic function testing laboratory after overnight fasting and were advised to wear loose-fitting clothes. They were instructed to avoid caffeine for 12 h before the test. The individuals were told to refrain from medications known to influence CV system, namely., anticholinergics, antihistamines, over-the-counter cough and cold medications, diuretics, and sympathomimetic and parasympathomimetic agents for 48 h before the study. Individuals were advised to discontinue short-acting α and β antagonists 24 h before and long-acting antagonists 48 h before the test. In case of any adversity in the health of the individuals resulting in poor overnight sleep quality, or any prodromal symptoms of fever and body pain, the test was postponed. The individuals were advised to come after evacuation of bowels and emptying of bladder. Height and weight were measured, and body mass index (BMI) was calculated for each subject.

Recording of short-term heart rate variability

The individuals were asked to lie comfortably on a couch and relax for 10 min. They were informed about the procedure to alleviate anxiety. Appropriate transducers were connected to monitor the electrocardiography (ECG) (lead II). The data acquisition was done at the rate of 500 samples per second for each channel using PowerLab 8/30 ML 870 Data Acquisition System with LabChart Pro software. Basal supine blood pressure (BP) and heart rate (HR) were recorded by oscillometric method using automated BP monitor Omron MX3. Following this, the lead II ECG was recorded for next 5 min in total resting condition for short-term HRV analysis, following the procedure as described earlier.[11] Power spectral analysis was done by fast Fourier transform and the frequency domain indices computed were very low frequency (LF), LF, and high frequency (HF); both in absolute powers given as ms^2 and in normalized unit (nu). The ratio of LF/HF in ms^2 was given as LF-HF ratio. The time domain measures included the various statistical measures from R-R interval (ms), standard deviation of normal to normal intervals (SDNN), root mean of standard deviations (RMSSD), normal to normal intervals (NN50) and pNN50.

Cardiovascular autonomic function tests

Heart rate response to standing

After 10 min of supine rest (from the basal recording measurements), the second recording of lead II ECG was started and the individuals were asked to stand after 30 s with a due marking on the graph. The postural change was obtained within 3 s.[12] HR were serially measured for next 5 min of the stand, i.e. immediate and 1st, 2nd, 3rd, 4th, and 5th min. Following this, the individuals were asked to sit down and allowed to rest for 3 min to achieve baseline HR values. RR tachogram from the ECG recording was retrieved and copied to a Microsoft Excel worksheet. A graphical representation (line diagram) of the RR tachogram was used to identify the minimum and maximum R-R interval after standing. The shortest R-R interval at or around the 15th beat and the longest R-R interval

at or around the 30th beat were calculated and thereby 30:15 ratio was computed.

Heart rate response to deep breathing

Deep breathing was performed at the rate of 6 breaths per minute with inspiratory and expiratory cycles for 5 s each.[12] During the 3 min of rest period after standing, deep breathing synchronized to a voice metronome and if necessary guided by hand movement was demonstrated to the individuals. The individuals performed deep breathing for the next 1 min with continuous lead II ECG recording. Individuals were encouraged to perform deep and maximal respiration. A period of 3 min rest was given after the maneuver for the HR and BP to return to basal state. RR tachogram from the corresponding ECG recording was retrieved and transformed graphically into a line diagram on a Microsoft Excel worksheet. The maximum and minimum R-R interval averaged over 6 cycles of expiration (E) and inspiration (I) was calculated and the ratio was computed as E: I ratio.

Blood pressure response to sustained isometric handgrip

Initially, the maximal voluntary contraction (MVC) during sustained isometric handgrip by the individuals was measured using handgrip dynamometer (Inco, Ambala). Then, the individuals were instructed regarding sustaining the handgrip at one-third of their MVC. The ECG recordings were started, and at the 15th s, individuals were instructed to perform one-third of their MVC for 3 min. The maximum diastolic BP (DBP) attained during the maneuver was noted. The difference between this highest DBP recorded during sustained handgrip and baseline supine DBP was noted as the pressor response to the test. After the procedure, BP and HR were measured after 2 min to confirm if they have returned to basal levels. The magnitude of DBP rise during the maneuver (ΔDBP_{isg}) was calculated as the difference between the maximum DBP attained during the handgrip and the supine basal values.

Recording of blood pressure variability parameter

The CV parameters and BRS were measured by continuous BPV method using Finapres (Finometer version 1.22a, Finapres Medical Systems BV, Amsterdam, The Netherlands), a noninvasive continuous hemodynamic CV monitor based on the principle of measurement of finger arterial pressure with the volume clamp technique of Penaz and the Physiocal criteria of Wesseling and as described earlier.[13] The reconstructed brachial pressure from finger pressure was acquired via a PC-based data acquisition system. Baroreflex sensitivity (BRS) was assessed along with other BPV parameters.

Estimation of biochemical parameters

Five milliliters of venous blood (after 12 h of overnight fasting) was collected from both cases and controls under aseptic precautions for biochemical analysis of following parameters.

Estimation thyroid and lipid profile and inflammatory marker

Free triiodothyronine (fT_3), free thyroxin (fT_4), thyroid-stimulating hormone (TSH), using standard chemiluminescence method with ADVIA Centaur (Siemens, Germany), were calculated. Total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), triglycerides (TG), low-density lipoprotein cholesterol (LDL-C), very LDL-C (VLDL-C), high sensitivity C-reactive protein (hs-CRP) were estimated. Lipid risk ratios such as TC/TG, TC/HDL-C, LDL/HDL-C, TG/HDL-C, and atherogenic index (AI) = log (TG/HDL − C) were calculated.

Statistical analysis of data

SPSS version 20 (20, Chicago, USA) was used for statistical analysis. The data were subjected to Kolmogorov–Smirnov normality test. All the data were expressed as mean ± SD. The intergroup differences in mean between the controls and cases were compared using Student's unpaired *t*-test for normally distributed data. Mann–Whitney test was used as the appropriate nonparametric test, for the parameters not following normal distribution. The association between LF-HF ratio with basal HR, BP, AFT parameters, and biochemical parameters was assessed by Pearson's correlation analysis and Spearman's correlation test as nonparametric test when appropriate. Multiple regression analysis was done to assess the independent contribution of BRS and TSH to LF-HF ratio and bivariate logistic regression analysis was done to demonstrate prediction of prehypertension status by BRS in SCH group. The difference was considered statistically significant if probability of chance was <0.05.

Results

As shown in Table 1, there was no significant difference in age, body weight, and BMI between control and study groups. Table 2 shows the comparison of frequency domain indices between cases and controls. On analysis of the frequency domain parameters of short-term HRV, total power (TP) was significantly reduced among the cases ($P < 0.0001$). When the absolute powers were expressed in nu, significantly elevated LFnu ($P = 0.0267$) and a depressed HFnu ($P = 0.0193$) were observed in cases in comparison to controls. The LF-HF ratio was significantly elevated in cases ($P = 0.0003$). Significant decrease was observed in NN50 ($P = 0.0047$), pNN50 ($P < 0.0001$), RMSSD ($P < 0.0001$), and SDNN ($P = 0.0258$) among cases. The pNN50 observed in cases was 62.22% lower than controls [Table 2].

Table 3 depicts the comparison of basal CV and continuous BPV parameters between the cases and controls. There was significant elevation of basal CV parameters i.e. systolic BP (SBP, $P = 0.0007$),

Table 1: Age and anthropometric parameters of control and subclinical hypothyroid groups

Parameters	Controls ($n = 32$)	Subclinical hypothyroids ($n = 32$)	*P*
Age (years)	28.688±7.350	29.094±7.476	0.8273
Body weight (kg)	58.844±12.366	54.938±11.242	0.1897
BMI (kg/m^2)	24.326±4.457	23.380±4.317	0.3918

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The $P<0.05$ was considered statistically significant. BMI: Body mass index, SD: Standard deviation

DBP ($P = 0.0011$) and mean arterial pressure (MAP, $P = 0.0004$) among the cases in comparison to the controls. Difference in HR was not significant between the groups ($P = 0.3085$). Among continuous BPV parameters, there was significant elevation of total peripheral resistance (TPR) ($P = 0.0238$) and decrease in BRS ($P < 0.0001$) among the cases. Stroke volume ($P = 0.842$) and cardiac output ($P = 0.7200$) were not significantly altered among the cases. Reduced left ventricular ejection time was observed in cases.

Table 4 shows the comparison of AFT parameters between controls and subclinical hypothyroid groups. There was a significant reduction in E: I ratio ($P = 0.0003$) among the cases. 30:15 ratio ($P = 0.0153$) and Δ DBP during isometric handgrip ($P = 0.0375$) were significantly elevated among the cases compared to controls.

Table 5 shows the comparison between the thyroid profile between control and subclinical hypothyroid subjects. Significantly elevated TSH values ($P = 0.0012$) was observed in cases while there was no significant difference in fT_3 and fT_4 levels when compared to controls. Table 6 depicts the comparison of lipid profile and lipid risk ratios between the cases and controls. TG, LDL-C, and VLDL-C were significantly higher among the cases ($P = 0.0173$, $P = 0.0246$, and $P = 0.0366$, respectively). Significant reduction in HDL-C ($P < 0.0001$) was observed in cases. TC did not show significant difference between the two groups but was higher in cases. The lipid risk ratios TC/HDL-C, TG/HDL-C, and LDL/HDL and AI were significantly higher among the cases ($P < 0.0001$). Table 6 compares the inflammatory marker hs-CRP between the cases and controls. hs-CRP was found to be significantly increased in cases compared to the controls ($P < 0.0001$).

Table 7 shows the correlation of LF-HF with the CV and lipid risk factor parameters. In the present study, there is strong correlation of LF-HF ratio with BRS ($r = 0.342$, $P = 0.015$) and TSH ($r = 0.325$, $P = 0.027$). There was also significant positive correlation of TC/HDL ($r = 0.344$, $P = 0.014$), TG/HDL ($r = 0.286$, $P = 0.046$) and AI ($r = 0.303$, $P = 0.030$); however,

Table 2: Frequency and time domain indices of heart rate variability of control and subclinical hypothyroid group

Parameters	Controls ($n=32$)	Subclinical hypothyroids ($n=32$)	*P*
TP (ms^2)	679.344±321.190	380.281±134.473	<0.0001***
LFnu	44.9±10.647	51.378±12.142	0.0267*
HFnu	55.1±9.479	48.622±11.952	0.0193*
LF-HF ratio	0.758±0.343	1.180±0.528	0.0003***
Mean RR (s)	0.818±0.122	0.774±0.090	0.057
RMSSD (ms)	49.18±20.606	30.588±14.007	<0.0001***
NN50	79.313±40.350	55.969±20.086	0.0047**
pNN50	39.022±17.031	14.741±7.577	<0.0001***
SDNN	46.188±23.870	33.469±20.550	0.0258*

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The $P<0.05$ was considered statistically significant. *$P<0.05$; **$P<0.01$; ***P<0.001. TP: Total power, LFnu: Low frequency component expressed as normalized unit, HFnu: High frequency component expressed as normalized unit, LF-HF ratio: Ratio of low-frequency power to high-frequency power of heart rate variability, RMSSD: Square root of the mean squared differences of successive normal to normal intervals, NN50: The number of interval differences of successive NN intervals >50 ms, pNN50: The proportion derived by dividing NN50 by the total number of NN intervals, SDNN: Standard deviation of normal to normal interval, SD: Standard deviation

Table 3: Continuous blood pressure variability parameters of control and subclinical hypothyroid group

Parameters	Controls ($n=32$)	Subclinical hypothyroids ($n=32$)	*P*
HR (/min)	76.340±9.990	78.733±8.601	0.3085
SBP (mmHg)	105.184±12.012	115.686±11.414	0.0007***
DBP (mmHg)	61.696±7.952	70.660±12.517	0.0011**
MAP (mmHg)	80.962±9.856	91.734±12.937	0.0004***
SV (ml)	73.094±13.523	73.912±18.899	0.842
LVET (ms)	326.696±15.427	319.110±20.294	0.0973
CO (L/min)	5.516±1.025	5.618±1.232	0.7200
TPR (mmHg/min/L)	0.902±0.139	1.093±0.445	0.0238*
BRS (ms/mmHg)	20.057±5.657	11.819±6.552	<0.0001***

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The $P<0.05$ was considered statistically significant. *$P<0.05$; **$P<0.01$; ***$P<0.001$. HR: Heart rate, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, MAP: Mean arterial pressure, SV: Stroke volume, LVET: Left ventricular ejection time, CO: Cardiac output, TPR: Total peripheral resistance, BRS: Baroreflex sensitivity, SD: Standard deviation

Table 4: Classical autonomic function testing parameters between control and subclinical hypothyroid groups

Parameters	Controls ($n=32$)	Subclinical hypothyroids ($n=32$)	*P*
30:15 ratio	1.347±0.150	1.464±0.219	0.0153*
E: I ratio	1.393±0.157	1.262±0.114	0.0003***
ΔDBP	16.188±6.567	19.375±5.36	0.0375*

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The $P<0.05$ was considered statistically significant. *$P<0.05$; **$P<0.01$; ***$P<0.001$. 30:15 ratio: Ratio between maximum RR interval at 30^{th} beat and minimum RR interval at 15^{th} beat, E: I ratio: Ratio of longest RR interval during expiration to the shortest RR interval during inspiration averaged over 6 cycles of respiration, ΔDBP: Difference in diastolic blood pressure between supine and isometric hand grip, SD: Standard deviation

Table 5: Thyroid profile between control and subclinical hypothyroid groups

Parameters	Controls ($n=32$)	Subclinical hypothyroids ($n=32$)	*P*
fT_3	2.942±0.408	2.987±0.616	0.7721
fT_4	1.038±0.173	1.006±0.285	0.5891
TSH	2.063±1.000	27.444±42.165	0.0012**

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The $P<0.05$ was considered statistically significant. *$P<0.05$; **$P<0.01$; ***$P<0.001$. fT_3: Free triiodothyronine, fT_4: Free thyroxin, TSH: Thyroid-stimulating hormone, SD: Standard deviation

Table 6: Lipid profile, lipid risk factors, and high-sensitive C-reactive protein between controls and subclinical hypothyroid groups

Parameters	Controls (n=32)	Subclinical hypothyroids (n=32)	P
Total cholesterol (mg/dL)	158.438±31.495	167.969±36.760	0.2697
Triglycerides (mg/dL)	83.125±24.874	97.938±23.561	0.0173*
HDL-cholesterol (mg/dL	45.594±11.821	34.656±8.40	<0.0001***
LDL-cholesterol (mg/dL)	95.900±29.358	113.725±32.475	0.0246*
VLDL-cholesterol (mg/dL)	16.913±5.300	19.588±4.7	0.0366*
TC/TG	2.031±0.603	1.779±0.419	0.056
TC/HDL	3.618±0.904	5.035±1.231	<0.0001***
TG/HDL	1.884±0.601	2.977±1.049	<0.0001***
LDL/HDL	2.219±0.826	3.44±1.096	<0.0001***
AI	0.254±0.140	0.451±0.142	<0.0001***
Hs-CRP (ng/mL)	826.644±327.7	1294.915±532.932	<0.0001***

Values expressed as mean±SD; analysis done by Student's unpaired *t*-test. The P<0.05 was considered statistically significant. *P<0.05; **P<0.01; ***P<0.001. HDL: High-density lipoprotein, LDL: Low-density lipoprotein, VLDL: Very low-density lipoprotein, TG: Triglycerides, AI: Atherogenic index, hs-CRP: High-sensitive C-reactive protein, TC: Total cholesterol, SD: Standard deviation

Table 7: Correlation of low-frequency to high-frequency with various parameters of control and subclinical hypothyroid subjects

Parameters	Controls (n=32)		Subclinical hypothyroids (n=32)	
	r	P	r	P
BMI	0.136	0.190	0.088	0.454
BHR	0.036	0.790	0.050	0.654
DBP	0.085	0.570	0.165	0.252
MAP	0.090	0.472	0.178	0.144
TPR	0.105	0.398	0.195	0.232
BRS	0.097	0.455	0.342	0.015*
TSH	−0.165	0.247	0.325	0.027*
TC/HDL	−0.084	0.565	0.344	0.014*
TG/HDL	−0.080	0.578	0.286	0.046*
LDL/HDL	−0.072	0.618	0.213	0.090
AI	−0.085	0.550	0.303	0.030*
hs-CRP	−0.202	0.163	0.205	0.094

The P<0.05 were considered statistically significant. BMI: Body mass index, BHR: Basal heart rate, DBP: Diastolic blood pressure, MAP: Mean arterial pressure, TC: Total cholesterol, TG: Triglycerides, HDL: High-density lipoproteins, LDL: Low-density lipoproteins, AI: Atherogenic index, TSH: Thyroid-stimulating hormone, hs-CRP: High-sensitive C-reactive protein, TPR: Total peripheral resistance, BRS: Baroreflex reflex sensitivity,*P < 0.05

there was no significant correlation of hs-CRP levels and with LF-HF ratio. Multiple regression analysis demonstrated significant independent contribution of BRS (B = 0.328, P = 0.012) and TSH (B = 0.356, P = 0.009) to LF-HF ratio [Table 8]. Furthermore, the bivariate logistic regression analysis demonstrated significant prediction of BRS by LF-HF ratio in SCH group (odds ratio [OR] = 2.17, P = 0.015) but not in control group (OR = 0.56, P = 0.175) [Table 9].

Discussion

The present study was designed to assess the CV functional status in the subclinical hypothyroid group and to correlate the nature and degree of alteration in autonomic status with the biochemical and metabolic derangements. The significant increase in LF-HF ratio in SCH group compared to the control group [Table 2] represents considerable SVI in subclinical hypothyroid patients as LH-HF ratio is a sensitive marker of sympathovagal balance.[10,14] Increase in LF-HF ratio in resting state indicates increased sympathetic activity and decreased vagal activity.[10,14] This was supported by significant increase in LFnu in SCH group compared to that of the control group [Table 2] as increased LFnu is an index of increased cardiac sympathetic drive.[10,14] In SCH subjects, significant decrease in HFnu indicates decreased parasympathetic tone in these patients as HFnu is an index of cardiac vagal drive.[10,14] This was supported by a significant reduction in TP of HRV in SCH group as TP in general indicates the magnitude of vagal modulation of cardiac function.[10,14] In addition, time domain indices of HRV (RMSSD, SDNN, NN50, and pNN50) were significantly less in SCH subjects [Table 2] further confirming decreased vagal tone in these patients, as time-domain indices of HRV represent cardiac parasympathetic drive.[10,14] Thus, it is evident from the present study that SVI in subclinical hypothyroid subjects is due to a concomitant increased sympathetic and decreased vagal activity.

In addition, there was altered autonomic reactivities in SCH subjects as evident from changes in 30:15 ratio, E: I ratio, and ΔDBP_{IHG} [Table 4]. HR response to standing (30:15 ratio) and deep breathing (E: I ratio) are parasympathetic function tests and BP response to isometric handgrip ($DDBP_{IHG}$) is sympathetic function test.[15] Significantly increased 30:15 ratio and decreased E: I ratio in SCH subjects compared to that of controls [Table 4] reflects lower vagal reactivity in subclinical hypothyroids.[16] A heightened diastolic pressure response to isometric handgrip ($DDBP_{IHG}$) in SCH subjects reflects increased sympathetic reactivity. This is consistent with the significantly high DBP in subjects of SCH group [Table 3] as increase in diastolic pressure in handgrip test depends primarily on vascular resistance that reflects sympathetic reactivity.[15] Thus, the findings of the present study substantiate that the

Table 8: Multiple regression analysis of low-frequency to high-frequency ratio (as dependent variable) with various other associated factors (as independent variables) in subclinical hypothyroid group

Independent variables	Standardized regression coefficient beta	95% CI of *B*		*P*
		Lower bound	Upper bound	
BRS	0.328	0.008	1.016	0.012*
AI	0.182	0.000	0.005	0.126
TSH	0.356	1.196	2.072	0.009**

The $P<0.05$ was considered statistically significant. BRS: Baroreceptor reflex sensitivity, TSH: Thyroid-stimulating hormone, AI: Atherogenic index, CI: Confidence interval,*$P < 0.05$, **$P < 0.01$

Table 9: Bivariate logistic regression analysis of prehypertension status (as dependent variable) with baroreceptor reflex sensitivity (as independent variable) in subclinical hypothyroidism group after adjusting for age, gender, and body mass index

OR	95% CI	*P*
2.17	1.118-4.216	0.015

The $P<0.05$ was considered statistically significant. CI: Confidence interval, OR: Odds ratio

SVI in SCH is due to both increased sympathetic activity and reactivity and decreased parasympathetic activity and reactivity.

Subclinical hypothyroid people have a myriad of factors which can affect the autonomic system and cause this alteration in the autonomic modulation in them. Hence, we have attempted to assess the contribution of inflammation and dyslipidemia in subclinical hypothyroid people to the autonomic dysfunction. Not only there were dyslipidemia (hypertriglyceridemia, high LDL-hypercholesterolemia, high VLDL-hypercholesterolemia and low HDL-cholesterolemia) and increased lipid risk factors [Table 6] in SCH group compared to that of control group, but also important lipid-related parameters were significantly correlated with LF-HF ratio [Table 7]. However, the degree of dyslipidemia in SCH group was mild as the levels of serum lipids were borderline high. Nevertheless, all the lipid risk factors except TC/TG were appreciably high in SCH group [Table 6]. Moreover, AI, the most important CV risk factor had significant correlation with LF-HF ratio [Table 7] though the direct contribution assessed by multiple regression analysis was not significant [Table 8]. From among the lipid risk factors assessed in the present study, we selected AI for the regression model as AI has recently been reported to be the better indicator CV risk.[17] Other lipid parameters were excluded from the same regression model to avoid multi-collinearity. Dyslipidemia is common in SCH,[18-20] and hyperlipidemia has been reported to be associated with increased sympathetic activity, especially in females.[21] Thus, from findings of the present study, it appears that chronic hyperlipidemia could be a major contributor to the SVI in SCH and AI may indirectly contribute to CV risk in this disorder.

There is report of low-grade immunological inflammation in subclinical hypothyroid subjects.[22] In the present study, marker of inflammation such as hs-CRP was significantly more in subjects of SCH group [Table 6]. However, hs-CRP was not significantly correlated with LF-HF ratio in SCH group [Table 7]. Thus, it appears that inflammation might not have a direct role to play in the genesis of SVI in SCH. Recently, it has been reported that CV disease risk in SCH is contributed by hs-CRP.[23] Therefore, we postulate that hs-CRP could be among possible links between SVI and CV risk in SCH, which needs further documentation in larger sample size. LF-HF ratio was significantly correlated with TSH in SCH group [Table 7], and TSH had independent contribution to LH-HF ratio [Table 8], indicating that the alteration in sympathovagal balance could be directly linked to the magnitude of SCH. However, further studies warrant the assessment of degree of SVI at different serum levels of TSH in subclinical hypothyroid patients to elucidate the association between autonomic imbalance and severity of SCH.

In the present study, SVI in subclinical hypothyroid subjects was associated with dyslipidemia, increased lipid risk factors, and inflammation that are known CV risk factors.[21,24] Moreover, the reduction in HRV *per se* has been reported to be an important CV risk.[25,26] In the present study, magnitude of HRV (represented by TP of HRV) was grossly reduced in subclinical hypothyroid patients compared to that of age- and BMI-matched control subjects [Table 2]. MAP was not significantly correlated with LF-HF ratio (SVI) in subjects of SCH group [Table 7]. As the age, gender, and BMI are known to influence SVI, the OR in logistic regression [Table 9] was calculated after adjusting for age, gender, and BMI (adjusted OR). SBP and DBP were significantly high in SCH group [Table 3]. Moreover, recently, we have reported the close association of SVI with increased vascular tone and hypertension status that increases CV risks in prehypertensives.[27] Therefore, SVI might contribute to rise in BP in SCH, which is linked to decreased BRS. The less sample size ($n = 32$) in SCH group might be the cause for lack of significant correlation of MAP with LF-HF ratio. A recent report has indicated that persistent hypertension in subclinical hypothyroid patients contributes to the cluster of CV disease risk factors.[28] Hence, it appears that SVI contributes to the CV risks in SCH. However, a study in larger sample size is warranted for elucidation of the link of SVI with hypertension status in SCH.

As there are reports of increased CV risks due to hyperlipidemia[18,29-31] and low-grade inflammation[4,19] in SCH patients, and in the present study, we found significant correlation between AI to SVI, we propose that a considerable decrease in HRV, intense SVI, persistent hyperlipidemia in SCH patients are interdependent phenomena. All these interlinked processes predispose these patients to increased

risk of CV in terms of morbidity and mortality and SVI could be the physiological basis of CV risks in SCH.

Arterial baroreceptors play a central role in BP regulation in response to various stimuli through alteration in both sympathetic and vagal activities, and therefore, assessment of baroreceptor reflex sensitivity (BRS) provides the state of autonomic imbalance in various CV disease states.[32] BRS has been documented as a measure of SVI and CV risk.[33] In the present study, BRS of SCH group was significantly reduced [Table 3] indicating attenuation of CV health of SCH patients as decreased BRS has been reported to be associated with CV morbidity.[33] Furthermore, it has been documented earlier that decrease in BRS predicts cardiac mortality in myocardial infarction patients.[34] The decrease in BRS was significantly correlated with LF-HF ratio [Table 7] and had significant independent association with LF-HF ratio [Table 8]. In addition, prediction of prehypertension by BRS was significant in SCH group [Table 9], as demonstrated by bivariate logistic regression. Thus, decreased BRS in SCH patient as observed in the present study could increase their CV risks and predispose them to adverse CV events. The present study is the first of its kind assessing the status of BRS with prehypertension status in SCH.

TPR was high in SCH group compared to control group [Table 3] though it was not significantly correlated with LF-HF ratio [Table 7]. TPR, an indicator of sympathetic vasoconstrictor tone, has been suggested as a predictor of CV disease risk in individuals who had more body size at birth.[35] In addition, TPR has been directly correlated with ventricular hypertrophy in hypertensives.[36] Thus, increase in TPR could also contribute cardiac morbidities in these subjects. In the present study, systolic prehypertension (SBP 120–139 mmHg) was observed in about 50% of SCH subjects, and decreased BRS had significant prediction of prehypertension status. Thus, it indicates that decreased BRS, which is a known CV risk, is closely linked to the development of prehypertension in SCH.

Conclusion

SVI and decreased BRS were observed in SCH group that was linked to SVI. Thus, it indicates that decreased BRS, which is a known CV risk, is closely linked to the development of prehypertension in SCH. Reduced BRS that contributes to prehypertension status predisposes to subclinical hypothyroids to CV risks.

Limitations of the study

The major limitation of the study is the modest number of sample size. In addition, we have not assessed the biochemical markers of sympathetic activity for assessment of SVI, and we have not done echocardiography for the assessment of cardiac dysfunctions in SCH subjects.

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Conflicts of interest

There are no conflicts of interest.

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