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Association between menopausal status and obesity and its effect on lipid profile in normal and hypertensive women.

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ABSTRACT

Present work is a continuation of our previous work where 78 pre- (39) and postmenopausal (39) women were recruited. Each group was further divided into; non-obese normal (10), obese normal (10), non-obese hypertensive (10), and obese hypertensive (10). Body mass index was calculated as per WHO and hypertensive status was determined following JNC (VII) guidelines. After ensuring fasting for 12 hours, blood sample of 5 ml was collected in sterile tubes from anti-cubital vein. Serum was separated, aliquots were made and serum samples were stored in freezer for the estimation of lipid profile and estradiol levels. Estradiol was assayed by competitive ELISA using Estradiol Kit manufactured by Adaltis Italia (Italy) for the previous work and rest of the sample was stored for assaying lipid profile for the present work. Lipid profile was estimated from the stored blood samples using the commercially available kits. The mean values of TC, TG and LDL-C were significantly higher whereas, mean value of HDL-C was significantly lower in obese normal subjects as compared to their non-obese counterparts in both pre- and postmenopausal groups. As compared to premenopausal, the situation was bad in postmenopausal women. Similar was the status in hypertensive subjects but the situation aggravated in hypertensive subjects as compared to their normal counterparts. The correlation exists between lipid profile and BMI and estradiol. Decreased levels of estradiol in obese and hypertensive subjects reflect the role of the hormone in making the situation worst. May be this is the reason, the prevalence of hypertension increases in obese as well as postmenopausal women.

Key words: lipid profile, hypertensive, menopausal status, premenopausal, postmenopausal, obese

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INTRODUCTION

Obesity, an increasingly prevalent and difficult-to-treat condition, affects more women than men. As per van der Merwe [1], latest prevalence figures for obesity within the European region confirm that in most countries, the number of obese women surpasses the number of obese men, sometimes being double than men. Many reports on obesity in India have also confirmed the prevalence of obesity to be higher among women than men [2,3, 4]. Obesity in itself has negative consequences for women's health throughout the life cycle. The growing prevalence of obesity is increasingly recognized as one of the most important risk factors for the development of hypertension, lipid abnormalities and type 2 diabetes mellitus (T2DM), which are known to be independent risk factors for cardiovascular diseases (CVDs) [5,6,7].

Hypertension has become a major health concern, possible a consequence of the rapid epidemiological transition over the last 2 or 3 decades. The prevalence of hypertension has been increasing, and in year 2000, an estimated 972

million people, out of which 333 million in economically developed and 639 million in economically developing countries, in the world were suffering from this problem. Compared with the year 2000, the number of people with hypertension in economically developed countries is projected to increase by 24% from 333 million to 413 million and a rise of 80% is predicted for economically developing countries from 639 million to 1·15 billion by the year 2025. On the basis of these estimates, almost three-quarters of the world's hypertensive population will be in economically developing countries by 2025 [8], Subjects with hypertension possess two folds higher risk of developing coronary artery disease, four times higher risk of congestive heart failure and seven times higher risk of CVD compared to normotensive subjects [9,10]. Although India is the second largest nation, relatively little is known about the actual prevalence of hypertension in India.

In females, weight gain particularly after menopause seems to be a universal phenomenon. Before the age of 50, majority of women tend to increase their weight slowly, whereas, after menopause there appears to be an accelerated increase in fat mass and a change in preferential fat storage to a central part of body that is abdominal location. Studies have reported that during menopause, weight gain and onset or worsening of obesity is favoured and prevalence of obesity is highest [11,12, 13 (a)]. It has been reported by Evans and Racette [14] that seventy percent women of age 45-54 years are over weight or obese. So, it may be concluded that the prevalence of obesity increases in postmenopausal women as compared to premenopausal women. According to Solimene [15], heart disease is the first killer of women in the modern era, regardless of age, race and of ethnicity, and its prevalence rises after menopause.

Menopause appears to be associated with adverse changes in blood lipid profile and these changes may speed up the process of atherosclerosis and especially coronary heart diseases which is the major cause of death and disability in postmenopausal women [16, 17]. Keeping all these factors in mind, the present study was designed to study the status of lipid profile in non-obese normal and hypertensive, obese normal as well as hypertensive pre- and postmenopausal women. The purpose was to observe if any relationship is established between menopausal status and the lipid profile.

MATERIALS AND METHODS

Present work is a continuation of our previous work where correlation between leptin and hypertension was established among non-obese and obese women with respect to their menopausal status [18]. To carry that work, 78 pre- (39) and postmenopausal (39) women were recruited. Each group was further divided into; non-obese normal (9), obese normal (10), non-obese hypertensive (10), and obese hypertensive (10). Here 'normal' stands for 'non-hypertensive'. The study was approved by the Guru Nanak Dev University Ethical Review Committee. Body mass index (BMI) was calculated as per WHO [19] and hypertensive status was determined following JNC VII [20] guidelines. While choosing the subjects for biochemical studies, it was ensured that all the pre- and postmenopausal subjects (in each subgroup) were having similar values of mean BMI so that the actual effect of their menopausal status could be ascertained. After ensuring fasting for 12 hours, blood sample of 5 ml was collected in sterile tubes from anti-cubital vein under aseptic conditions from the selected non-obese and obese (Normal and hypertensive) pre- and postmenopausal subjects who consented for blood sample collection. Blood samples were allowed to stand and then centrifuged. Serum was separated, aliquots were made and serum samples were stored in freezer for the estimation of lipid profile and estradiol levels. Estradiol was assayed by competitive ELISA using Estradiol Kit manufactured by Adaltis Italia (Italy) for the previous work and rest of the sample was stored for assaying lipid profile for the present work.

Lipid profile was estimated from the stored blood samples using the commercially available kits. Total serum Cholesterol (TC) was determined by enzymatic (CHOD-PAP) colorimetric method [21] and Triglyceride (TG) by enzymatic (GPO-PAP) method [22]. High density lipoprotein (HDL-C) was estimated by precipitation method [23] and Low density lipoproteins (LDL-C) by Friedewald formula [24].

Data analysis

Data was maintained on excel spread sheet. Analysis was performed using SPSS (Statistical Package for Social Sciences, SPSS Inc., Chicago, IL, USA)) version 16 for windows. Results were presented as mean± S.D. The differences in biochemical variables between Pre-M and Post-M women were assessed with't-test'. ANOVA was done to analyze the comparison between the groups.

RESULTS

Estradiol levels were assessed among the same sample groups during our previous study. It was observed that the level of estradiol was significantly lower in obese (normal as well as hypertensive) premenopausal women as compared to non-obese premenopausal subjects. Similar trend of change was observed in postmenopausal group. On comparing non-obese pre- and postmenopausal women, significantly lower mean values of estradiol were observed in postmenopausal women in both the groups; normal as well as hypertensive ($p<0.001$). Similar status was observed in the obese pre- and postmenopausal women showing significantly lower levels of estradiol in postmenopausal women ($p<0.001$) in both the groups as compared to their premenopausal counterparts (data not shown).

Lipid Profile of Normal and Hypertensive Pre- and Postmenopausal subjects

The comparative account of lipid profile of the normal pre- and postmenopausal subjects is given in **Table 1**. In normal subjects, the mean values of TC, TG and LDL-C were significantly higher ($p<0.001$, $p<0.05$; $p<0.001$, respectively), whereas, mean value of HDL-C was significantly lower ($p<0.02$) in obese normal subjects as compared to their non-obese premenopausal counterparts. Similar trend of changes in lipid profile was observed in normal postmenopausal women (**Table 1**). Levels of TC, TG and LDL-C were found statistically higher in obese subjects ($p<0.02$) as compared to their non-obese counterparts.

Table 1: Lipid Profile of Normal Non-Obese and Obese Premenopausal and Postmenopausal Women

Component	Premenopausal Women		Postmenopausal Women	
	Non-Obese	Obese	Non-Obese	Obese
TC (mg/dl)	152.59±20.76	201.57±30.28^{d}	168.21±17.50$^{a''}$	205.70±33.88$^{b'}$
TG (mg/dl)	90.12±17.98	131.20±49.70^{a}	107.09±29.26	142.67±18.20$^{b'}$
LDL-C (mg/dl)	73.17±12.13	135.89±25.54^{d}	104.82±20.69$^{c''}$	146.71±32.75$^{b'}$
HDL-C (mg/dl)	62.03±13.44	43.15±9.71^{b}	46.37±5.02 $^{d''}$	40.48±10.62

The values are Mean± SD. (a), (b), (d) represents the comparison between non-obese and obese premenopausal women; (b') stands for comparison between non-obese and obese postmenopausal women; (a''), (c''), (d'') compares the mean values of non-obese pre- and postmenopausal women, where a, a''=p<0.05, b, b'=p<0.02, c''=p<0.01, d, d''=p<0.001

On comparison between normal pre- and postmenopausal subjects, the levels of TC, and LDL-C were observed higher in both, non-obese and obese postmenopausal women as compared to non-obese and obese premenopausal subjects with statistically significant increase in non-obese group only (TC: $p<0.05$, LDL-C: $p<0.0.1$). On the other hand, HDL-C content showed decline in both non-obese and obese postmenopausal subjects as compared to premenopausal women with statistically significant difference in non-obese group ($p<0.001$).

The mean values of lipid profile in hypertensive non-obese and obese pre- and postmenopausal women are presented in **Table 2**. It is evident from these values that TC, TG and LDL-C levels were significantly higher in obese hypertensive women as compared to non-obese pre- and postmenopausal women ($p<0.02$ in TC, $p< 0.01$ in TG and LDL-C in pre and $p<0.02$ in TC, TG and $p<0.01$ in LDL-C in postmenopausal women). On the other hand, lower mean values of HDL-C were observed in obese hypertensive pre- and postmenopausal women as compare to their normal counterparts but the differences were statistically non-significant.

Table 2: Lipid Profile of Hypertensive Non-Obese and Obese Premenopausal and Postmenopausal Women

Component	Premenopausal Women		Postmenopausal Women	
	Non-Obese	Obese	Non-Obese	Obese
TC (mg/dl)	175.76±18.08	206.97±26.27^{b}	181.33±7.46	211.28±31.23$^{b'}$
TG (mg/dl)	100.73±15.83	133.42±23.42^{c}	125.63±13.10$^{c''}$	143.63±9.57$^{b'}$
LDL-C (mg/dl)	110.04±17.95	137.87±12.54^{c}	118.56±11.90	148.20±15.64$^{c'}$
HDL-C(mg/dl)	43.03±5.55	40.83±8.65	36.66±4.14$^{c''}$	32.91±5.11$^{a'''}$

Values are Mean ±SD. (b),and (c) stands for comparison between non-obese and obese premenopausal women; (b'),(c') stands for comparison between non-obese and obese postmenopausal women; (c'') compares the mean values of non-obese pre- and postmenopausal women and (a''') compares the mean values of obese pre- and postmenopausal women where a'''=p<0.05, b, b'=p<0.02, c, c', c''=p<0.01

When compared the mean values of lipid profile between hypertensive pre- and postmenopausal women, higher mean values of TC, TG, and LDL-C were observed in non-obese postmenopausal women but the difference was statistically significant in case of TG only ($p<0.01$), whereas, significantly lower HDL-C level ($p<0.01$) was observed in hypertensive non-obese postmenopausal subjects as compared to their premenopausal counterparts.

Similarly, on comparison between obese women of both the groups, TC, TG and LDL-C levels showed higher mean values in obese postmenopausal women as compared to their obese premenopausal counterparts but the differences were not statistically significant. On the other hand, significantly lower level of HDL-C ($p<0.05$) was observed in obese postmenopausal subjects as compared to obese premenopausal women.

Table 3: Pearson's correlation co-efficient (r) of TC, TG, LDL-C and HDL-C with different variables in Normal Non-Obese and Obese Premenopausal and Postmenopausal Women

Variables	TC (mg/dl)		TG (mg/dl)		LDL-C (mg/dl)		HDL-C (mg/dl)	
	Non-Obese	Obese	Non-Obese	Obese	Non-Obese	Obese	Non-Obese	Obese
Premenopausal Women								
BMI (kg)/(m^2)	0.576	0.608	0.536	0.870***	0.404	0.831***	-0.804**	-0.931***
SBP (mm/Hg)	0.262	0.556	0.139	0.553	0.480	0.440	0.030	-0.598
DBP (mm/Hg)	0.372	0.501	0.619	0.622	0.420	0.544	-0.544	-0.614
Estradiol(pg/ml)	-0.723*	-0.732*	-0.449	-0.627	-0.724*	-0.723*	0.704*	0.723*
Postmenopausal Women								
BMI (kg)/ (m^2)	0.683*	0.675*	0.752*	0.501	0.782*	0.373	-0.737*	-0.359
SBP (mm/Hg)	0.350	0.856***	0.003	0.140	0.026	0.006	-0.127	-0.496
DBP (mm/Hg)	0.655	0.379	0.448	0.743	0.325	0.590	-0.345	-0.786*
Estradiol(pg/ml)	-0.882**	-0.833**	-0.452	-0.727*	-0.798**	-0.746*	0.722*	0.756*

*BMI-Body mass index, SBP-Systolic blood pressure, DBP-Diastolic blood pressure, where p≤0.001=***, p≤0.01=**, p≤0.05=**

Correlation of Lipid Profile with different variables

Tables 3 and 4 depict the correlation of lipid profile with different variables in normal as well as hypertensive subjects. The table **3** shows positive correlation of TC, TG, LDL-C with BMI, SBP, DBP and negative correlation with estradiol, whereas, HDL-C shows negative correlation with BMI, SBP, DBP and positive correlation with estradiol among normal pre- and postmenopausal women. Among premenopausal women, correlation of lipid profile is stronger with BMI and estradiol, whereas, among postmenopausal women, TC is strongly correlated with SBP and HDL-C with DBP among obese subjects besides all other components of lipid profile being strongly correlated with BMI and estradiol. It is further reflected that this correlation is stronger in postmenopausal women as compare to premenopausal subjects.

Table 4: Pearson's correlation co-efficient (r) of TC, TG, LDL-C and HDL-C with different variables in Hypertensive Non-Obese and Obese Premenopausal and Postmenopausal Women

Variables	TC (mg/dl)		TG (mg/dl)		LDL-C (mg/dl)		HDL-C (mg/dl)	
	Non-Obese	Obese	Non-Obese	Obese	Non-Obese	Obese	Non-Obese	Obese
Premenopausal Women								
BMI (kg)/(m^2)	0.776**	0.869***	0.961***	0.685*	0.924***	0.909***	-0.721*	-637*
SBP (mm/Hg)	0.822**	0.818**	0.863***	0.939***	0.612	0.681*	-0.704*	-0.787**
DBP (mm/Hg)	0.943**	0.923***	0.839**	0.742**	0.844**	0.834**	-0.840**	-0.518
Estradiol(pg/ml)	-0.758**	-0.702*	-0.492	-0.754*	-0.800**	-0.499	0.710*	0.802**
Postmenopausal Women								
BMI (kg)/(m^2)	0.757*	0.731*	0.885***	0.687*	0.826**	0.218	-0.739*	-0.739*
SBP (mmHg)	0.714*	0.632*	0.879***	0.602	0.964***	0.702*	-0.667*	-0.153
DBP (mmHg)	0.780*	0.836**	0.872***	0.627*	0.798**	0.714*	-0.787**	-0.487
Estradiol(pg/ml)	-0.823**	-0.802**	-0.827**	-0.707*	-0.808**	-0.506	0.724*	0.832**

*BMI-Body mass index, SBP-Systolic blood pressure, DBP-Diastolic blood pressure, where, p≤0.001=***, p≤0.01=**, p≤0.05=**

Among hypertensive subjects (**Table 4**), TC, TG, LDL-C is significantly positively correlated with BMI, SBP, DBP and negatively correlated with estradiol, whereas, HDL-C is significantly negatively correlated with BMI, SBP, DBP and positively correlated with estradiol level. It is further reflected that this association is stronger among postmenopausal women as compared to their premenopausal counterparts.

DISCUSSION

Biochemical analysis revealed significant differences in the lipid profile of non-obese and obese pre- and postmenopausal women in both the studied groups. The TC, TG, LDL-C levels were significantly higher while HDL-C levels were significantly lower in obese subjects as compared to non-obese in both, pre- and postmenopausal women. Results of the present study are supported by the findings of other researchers. Chadha et al [25] in their study on correlation of lipid profile with anthropometric variables observed positive correlation of elevated cholesterol and LDL-C with BMI. Sandhu et al. [26] in their study observed positive correlation between

TG and BMI and negative correlation between HDL-C and BMI. It is evident from these reports that with increase in weight and BMI, the mean values of non-friendly components of lipid profile increase and friendly HDL-C decrease. Both, TC and TG levels reflect free fatty acids in plasma. The level of fatty acids is elevated in obesity because greater than normal amount of free fatty acids are released from increased adipose tissue mass even though the fatty acid release from individual cell is normal [27]. On comparison between pre- and postmenopausal women, higher mean values of TC, TG, LDL-C and lower HDL-C were observed in non-obese and obese postmenopausal women as compared to premenopausal women. It may be attributed to the decline in estradiol level at menopause in the postmenopausal women, as withdrawal of estrogen leads to poor lipid profile in women. 10-20% increase in TC, and LDL-C and 10% decrease in HDL-C level after 2 years of menopause has been reported [28]. Fall in estrogen level at menopause deteriorates the lipid profile [29]. As estrogens are associated with lowering cholesterol, and in particular reducing LDL, the fall in their level at menopause may be attributed to higher rate of obesity among postmenopausal women [30].

The levels of different components of lipid profile play a significant role in determining the health of an individual. In the hypertensive subjects, lipid profile got worsened. Higher levels of TC, TG, LDL-C and lower levels of HDL-C were observed in hypertensive non-obese and obese pre- as well as postmenopausal women as compared to normal subjects. Our findings are in agreement with the findings of other scientists who observed higher levels of TC, TG, LDL-C and lower levels of HDL-C among hypertensive as compared to normotensive subjects [31, 32]. Liver plays an important role in lipid metabolism. It is the hub of fatty acid synthesis and lipid circulation through lipoprotein synthesis [33]. Estradiol has an impact on the liver. Estrogen increases the activity of lipase hepatic enzymes and, consequently, the levels of TC, TG and LDL-C decrease and the synthesis of HDL-C is stimulated [34, 35]. These findings suggest some link among dyslipidemia, estradiol and hypertension that might be relevant to the development of CVD in obese subjects.

Improved levels of lipid profile and insulin resistance after administration of estrogen among postmenopausal women further supports the relationship between reproductive hormones and lipid profile [36]. As the situation becomes unfavourable in postmenopausal women as compared to premenopausal women in both normal as well hypertensive subjects, it is suggested that declined estradial in postmenopausal women plays significant role in making the situation worse. So, postmenopausal women must be advised to take care of their life style, eat healthy food and do regular work out.

CONCLUSION

Decreased levels of estradiol in obese and hypertensive subjects reflect the role of the hormone in making the situation worst among these subjects. May be this is the reason, the prevalence of hypertension increases in obese as compared to non-obese as well as in postmenopausal women as compared to their premenopausal counterparts..

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