Original Research Article

To study the carotid intima media thickness in patients of fatty liver disease

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ABSTRACT

Background: Fatty liver is associated with several atherosclerotic risk factors such as hypertension, diabetes and dyslipidemia. It has also been related to insulin resistance. This association was found in NIDDM patients as well as in non-diabetic subjects. An increased intima-media thickness (IMT) has been shown to be a risk factor for myocardial infarction and stroke. The aim of the present study is to investigate associations between hepatic steatosis and the risk of atherosclerosis.

Methods: The present study was carried out on 88 patients of fatty liver disease and 80 controls in the department of General Medicine. An approximate equal number of age and sex matched persons without fatty liver were selected randomly as controls. Both fatty liver disease patients (i.e NAFLD and AFLD) and control group were further divided into two categories, one with risk factor for atherosclerosis and other without risk factors. Risk factors for atherosclerosis were taken according to ATP III guidelines.

Results: When comparison of mean CIMT was done in NAFLD, AFLD and controls in a particular age group, significant difference was found in mean CIMT (both sides) in age group 40–49 yrs (p value 0.03, 0.002 for right and left respectively). The difference was also significant in mean CIMT of right in age group 18–24 yrs (p value 0.015) and in >60 years (p value 0.03). Among, NAFLD patients, for left mean CIMT p value was 0.0001, for right mean CIMT p value was 0.0001. Among AFLD patients, for left mean CIMT p value was 0.006 and for right mean CIMT p value was 0.0022. Only statistically significant difference was found in mean CIMT (left) in grade I fatty liver (p value 0.04). NAFLD and controls without risk factors for atherosclerosis, mean CIMT (both side) in AFLD patients was found to be significantly more than control (p value 0.04). AFLD patients and controls without risk factors for atherosclerosis, mean CIMT of both side in AFLD patients was found to be significantly more than controls (p value for left CIMT 0.02 and for right CIMT 0.0001).

Conclusions: CIMT was found to increase with advancing age in all three group i.e. NAFLD, AFLD and control group. CIMT was more in patients of fatty liver disease (both NAFLD and AFLD) having risk factor for atherosclerosis as compared to those without risk factors. Both NAFLD and AFLD are associated with increased CIMT in comparison to control group. As such all NAFLD and AFLD patients should be investigated for carotid atherosclerosis, as its early detection and management may be helpful in limiting the inherent complications of atherosclerosis.

Keywords: AFLD, IMT, NIDDM, NAFLD

INTRODUCTION

Fatty liver is a common clinical and histological finding. Fatty liver can be non-alcoholic fatty liver disease (NAFLD) or alcoholic fatty liver disease (AFLD) broadly. Non-alcoholic fatty liver disease (NAFLD) is defined as a fatty liver (liver fat >5-10% of liver weight),
which is not due to excess alcohol consumption or other causes of steatosis.¹

NAFLD is the most common cause of elevated liver function tests (LFTs) in the US according to the (NHANES III) survey.² If there is no co-existing liver disorder such as alcoholic hepatitis or steatohepatitis, further local organ damage does not occur.³ In the clinic, it is difficult to distinguish between subjects with NAFLD and AFLD using abnormal liver function tests (LFTs).

Fatty liver is associated with several atherosclerotic risk factors such as hypertension, diabetes and dyslipidemia. It has also been related to insulin resistance.⁴ This association was found in NIDDM patients as well as in non-diabetic subjects.⁵ Although an association between hepatic steatosis and atherosclerotic risk factors has been described, possible direct relationships between hepatic steatosis and atherosclerosis remain to be investigated.

The intima-media thickness (IMT) of the carotid artery can be measured non-invasively by ultrasound techniques. An increased IMT has been shown to be a risk factor for myocardial infarction and stroke.⁶ Furthermore, carotid ultrasound is an accurate diagnostic tool for detecting atherosclerotic plaques and for assessing the degree of luminal narrowing caused by atherosclerotic changes of the vessel wall.⁷

**METHODS**

The present study was carried out on 88 patients of fatty liver disease and 80 controls over a period of one year in the department of General Medicine, G.R. Medical College, Gwalior, Madhya Pradesh. On the basis of history of alcohol intake, patients consuming ≤ 20 g (for men) and ≤ 10 g (for women) of ethanol per day were considered to have NAFLD, while those consuming >20 g (for men) and >10 g (for women) of ethanol per day were considered to have AFLD. An approximate equal number of age and sex matched persons without fatty liver were selected randomly as controls. Both fatty liver disease patients (i.e NAFLD and AFLD) and control group were further divided into two categories, one with risk factor for atherosclerosis (Age- men >45 years; women >55 years, cigarette smoking, hypertension, diabetes mellitus, low HDL cholesterol (< 40 mg/dl), Obesity (BMI > 30kg/m²), atherogenic diet) and other without risk factors. Diagnosis of fatty liver disease was made on the basis of presence of fatty liver on abdominal ultrasonographic examination and graded as follows:

Grade 1: slight diffuse increase in the fine echoes. Liver appears bright compared to the cortex of the kidney. Normal visualization of diaphragm and intra hepatic vessel borders.

Grade 2: moderate diffuse increase in the fine echoes. Slightly impaired visualization of the intra hepatic vessels and diaphragm.

Grade 3: marked increase in the fine echoes. Poor or no visualization of intra hepatic vessel borders, diaphragm and the vessels.

Patients with conditions likely to alter serum aminotransferase levels like viral, autoimmune or toxic hepatic diseases, use of hepatotoxic drugs within six months, cases of surgery like gastrointestinal surgery, biliary obstruction and primary biliary cirrhosis and pregnancy were excluded from the study.

Informed consent was obtained from the patient or from a close relative. Cases and controls were subjected to a detailed history and thorough clinical examination as per proforma. Complete haemogram, ESR, urine R/M, FBS, PPBS, BUN, creatinine, liver function tests, lipid profile, HBsAg, fundus Examination, ECG, ultrasound Examination of abdomen and carotid Doppler were done in all.

Carotid intima media thickness was evaluated as a marker of atherosclerosis. The carotid arteries were bilaterally examined with high resolution B-mode ultrasound using a 5 MHz linear array transducer. The mean thickness of carotid intima media of > 1mm was defined as presence of carotid atherosclerosis. The data thus obtained was subjected to standard statistical analysis.

**RESULTS**

In the present study number of patients having NAFLD were 46 (52.27%) and those having AFLD were 42 (47.72%). Patients in the control group were 80, out of which 65% were male and 35% were female. More than 50% of the cases of AFLD were having grade II fatty liver (54.76%) while 25 out of 46 (54.34%) of NAFLD cases were having grade I fatty liver.

Maximum percentage of NAFLD cases were in age group 40-49 years. 60.8% were males while female constituted only 39.13%. The mean age of male cases was 48.07±13.37 years (range 24-71 years) and that of female cases was 43.83±17.75 years (range 19-76 years).

Maximum percentage of AFLD cases were in age group 18-29 years followed by 40-49 years, and all cases were males. The control cases were those without having fatty liver and approximately equally distributed in various age group. Out of these, 65% were males and 35% were females. The mean age of male cases in control group was 44±17.35 yrs. (range 19-82 years) and that of female patients was 45.14±12.49 years (range 20-70 years).

Increase in triglyceride level was found in both males and females having NAFLD (25% and 27.27% respectively). 46% of males and 61.11% of females with NAFLD were
having decreased HDL level indicating that dyslipidemia is associated with NAFLD. The most remarkable dyslipidemic change seen in AFLD cases in our study was decrease in HDL cholesterol level [13 out of 42 cases (30.95%)] followed by increase in triglyceride level [9 out of 42 (21.42%)].

Table 1: Comparison of CIMT in patients of NAFLD, AFLD and control group.

| CIMT (mm) | NAFLD (n=46) | AFLD (n=42) | Control (n=80) |
|-----------|--------------|-------------|---------------|
|           | M (n=28) | F (n=18) | P value | M (n=42) | F (n=0) | P value | M (n=52) | F (n=28) | P value |
| Mean CIMT (lt.) | 0.71±0.18  | 0.70±0.19  | 0.85  | 0.71±0.11 | - | - | 0.66±0.12  | 0.62±0.13  | 0.28  |
| Mean CIMT (rt.) | 0.72±0.17  | 0.70±0.18  | 0.70  | 0.71±0.16 | - | - | 0.65±0.14  | 0.66±0.14  | 0.76  |

Table 2: Age wise comparison of mean CIMT in NAFLD, AFLD and control group.

| Group | 18-29 (Years) | 30-39 (Years) | 40-49 (Years) | 50-59 (Years) | 60 and above (Years) | Anova P value | DOF | WITH IN | BETWEEN |
|-------|---------------|---------------|---------------|---------------|----------------------|----------------|------|---------|---------|
| Mean CIMT (mm) lt. | NAFLD (n=46) | 0.51±0.19 | 0.68±0.09 | 0.65±0.10 | 0.78±0.16 | 0.86±0.17 | 0.269 | 4 | 41 |
| AFLD (n=42) | 0.66±0.10 | 0.66±0.13 | 0.79±0.11 | 0.70±0.10 | 0.75±0.09 | 0.0384 | 4 | 37 |
| Control (n=80) | 0.55±0.09 | 0.63±0.13 | 0.64±0.11 | 0.69±0.14 | 0.73±0.11 | 0.00007 | 4 | 75 |
| Mean CIMT (mm) rt. | NAFLD (n=46) | 0.52±0.17 | 0.66±0.12 | 0.68±0.13 | 0.82±0.17 | 0.82±0.13 | 0.0005 | 4 | 41 |
| AFLD (n=42) | 0.65±0.16 | 0.68±0.16 | 0.82±0.19 | 0.63±0.05 | 0.75±0.13 | 0.108 | 4 | 37 |
| Control (n=80) | 0.55±0.10 | 0.63±0.15 | 0.68±0.10 | 0.68±0.16 | 0.73±0.13 | 0.003 | 4 | 79 |

Table 3: Comparison of mean CIMT in NAFLD, AFLD and control group according to age.

| Group | 18-29 (Years) | 30-39 (Years) | 40-49 (Years) | 50-59 (Years) | 60 and above (Years) |
|-------|---------------|---------------|---------------|---------------|----------------------|
| CIMT lt. | NAFLD (n=46) | 0.52±0.17 | 0.66±0.12 | 0.68±0.13 | 0.68±0.03 | 0.82±0.17 | 0.78±0.10 | 0.82±0.13 | 0.86±0.17 |
| AFLD (n=42) | 0.65±0.16 | 0.66±0.16 | 0.66±0.13 | 0.82±0.12 | 0.79±0.11 | 0.75±0.09 | 0.0384 | 4 | 37 |
| Control (n=80) | 0.55±0.10 | 0.63±0.15 | 0.68±0.10 | 0.68±0.16 | 0.73±0.13 | 0.003 | 4 | 79 |
| CIMT rt. | NAFLD (n=46) | 0.52±0.17 | 0.66±0.12 | 0.68±0.13 | 0.68±0.03 | 0.82±0.17 | 0.78±0.10 | 0.82±0.13 | 0.86±0.17 |
| AFLD (n=42) | 0.65±0.16 | 0.66±0.16 | 0.66±0.13 | 0.82±0.12 | 0.79±0.11 | 0.75±0.09 | 0.0384 | 4 | 37 |
| Control (n=80) | 0.55±0.10 | 0.63±0.15 | 0.68±0.10 | 0.68±0.16 | 0.73±0.13 | 0.003 | 4 | 79 |
| (ANOVA) P value | 0.08 | 0.015 | 0.66 | 0.68 | 0.03 | 0.002 | 0.107 | 0.37 | 0.19 | 0.03 |
| DOF (within) | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 |
| (between) | 33 | 33 | 34 | 34 | 38 | 38 | 25 | 25 | 33 | 33 |

CIMT was found to increase with age in all three groups. In control group, statistically significant difference was found in mean CIMT (both right and left) in different age groups, while in NAFLD significant difference was found in mean CIMT of right side and in AFLD difference was significant in left side as per age.

Significant difference was found in mean CIMT (both right and left) in age group 40-49 years. between...
NAFLD, AFLD and control group. The difference was also significant in mean CIMT of left side in age group 18-29 years and also in age group more than 60 years. Mean CIMT of both sides increased with increasing grades of fatty liver and was statistically significant in both sub groups.

Table 4: Comparison of mean CIMT between NAFLD and control group with and without risk factor for atherosclerosis.

| CIMT (mm) | Risk factor absent (n=64) | Risk factor present (n=62) |
|-----------|---------------------------|---------------------------|
|           | NAFLD (n=24) | Control (n=40) | P | NAFLD (n=22) | Control (n=40) | P |
| Mean CIMT (lt.) | 0.66±0.16 | 0.59±0.11 | 0.04 | 0.73±0.16 | 0.73±0.13 | 0.29 |
| Mean CIMT (rt.) | 0.65±0.18 | 0.58±0.10 | 0.04 | 0.76±0.17 | 0.72±0.12 | 0.28 |

Table 5: Comparison of mean CIMT between AFLD and control group with and without risk factor for atherosclerosis.

| CIMT (mm) | Risk factor absent (n=61) | Risk factor present (n=61) |
|-----------|---------------------------|---------------------------|
|           | AFLD (n=21) | Control (n=40) | P | AFLD (n=21) | Control (n=40) | P |
| Mean CIMT (lt.) | 0.66±0.11 | 0.59±0.11 | 0.02 | 0.76±0.19 | 0.73±0.13 | 0.469 |
| Mean CIMT (rt.) | 0.69±0.10 | 0.58±0.10 | 0.0001 | 0.73±0.12 | 0.72±0.12 | 0.758 |

Table 6: Comparison of mean CIMT in patients of fatty liver with and without risk factors (RF) for atherosclerosis.

| CIMT (mm) | NAFLD (n=46) | AFLD (n=42) |
|-----------|--------------|-------------|
|           | RF absent (n=24) | RF present (n=22) | P | RF absent (n=21) | RF present (n=21) | P |
| Mean CIMT (lt.) | 0.66±0.16 | 0.73±0.16 | 0.14 | 0.66±0.11 | 0.76±0.19 | 0.043 |
| Mean CIMT (rt.) | 0.65±0.18 | 0.76±0.17 | 0.03 | 0.69±0.10 | 0.73±0.12 | 0.24 |

No statistically significant difference in mean CIMT was found between NAFLD and AFLD group in all grades of fatty liver except in grade II fatty liver on left side where difference was statistically significant (p value 0.04). On comparison of mean CIMT between NAFLD and control without the risk factors for atherosclerosis the mean CIMT in NAFLD was found to be significantly more (p value 0.04), indicating that NAFLD per se is associated with increase in CIMT (atherosclerosis). The mean CIMT was significantly more in AFLD group in comparison to control group when risk factors for atherosclerosis were not present (p value is 0.02, 0.001 for mean CIMT (Lt.) and (Rt.) common carotid artery respectively). In the presence of risk factors for atherosclerosis, when NAFLD group was compared with control group and similarly AFLD group was compared with control group, there was no statistically significant difference.

Mean CIMT (right) in NAFLD patients with risk factors was found to be significantly (p 0.03) more than patients without risk factors. Mean CIMT (left) in AFLD patients with risk factors was found to be significantly (p 0.043) more than patients without risk factor.

**DISCUSSION**

Although the world-wide prevalence of NAFLD has not yet been determined, it is 10-24% in common population and 57.5% in obese individuals. It was reported in Japan that incidence of NAFLD in children with obesity is around 22.5% to 52.8%. Prevalence of NAFLD and AFLD in a study by Kotronen et al, was 21% and 7% respectively. In this study out of 88 cases of fatty liver, 46 (52.27%) were NAFLD and 42 (47.72%) were AFLD.

Agarwal R reported that 48.1%, 40.3% and 11.3% had grade I, II and III fatty liver respectively.10 In our study, in NAFLD group 25 (54.34%) patients had grade I fatty liver, 19 (41.30%) had grade II fatty liver and 2 (4.34%) had grade III fatty liver. Results in our study was comparable to the observation in the study mentioned above. In AFLD group 17 (40.47%) patients had grade I fatty liver, 23 (54.76%) patients had grade II fatty liver and 2 (4.7%) had grade III fatty liver.

Mean age of NAFLD patients in a study by Bacon et al was 47 years.11 Ludwig et al similarly noted mean patients age of 54 years.12 In various other studies mean age varied between 47 and 54 years. In our study, maximum number of subjects were in 5th decade. The age range of patients was from 18-82 years and the mean age was 46.41±15.18 years. Mean age in male patients was 48.07±13.37 years (range 24-71 years), and female patients was 43.83±17.75 years (range 14-76 years). Our results are in agreement with findings of others. In AFLD, maximum number of patients were in 3rd decade followed by 5th decade in our study.
Recent studies by Matteoni et al, George et al have shown that it occurs with equal frequency in men (approx. 50%), in addition even higher involvement of men had been noted. In the present study male preponderance was seen i.e. males constitute 60.8% of total and male:female ratio was 3:2. Women exhibit increases susceptibility to alcoholic liver disease (ALD) at amounts >20gm/day. Asian Indians are more commonly predisposed to atherogenic dyslipidemia (i.e. combination) of hypertriglyceridemia, low levels of HDL cholesterol and high levels of LDL cholesterol. This factor along with higher incidence of insulin resistance make a very fertile ground in Indians for having metabolic syndrome and its variable manifestation including NAFLD.

In this study in NAFLD patients mean CIMT of left common carotid artery in males was 0.71±0.18 mm and in females was 0.70±0.19 mm. Similarly, mean CIMT of right common carotid artery in males was 0.72±0.17 mm and females was 0.70±0.18 mm. This showed that there was no significant difference in mean CIMT between males and females of NAFLD group. No female was present in AFLD group so comparison was not done.

In our study mean CIMT was found to increase with increasing age in all the groups. In our study, in control group statistically significant difference was found in mean CIMT (both right and left side) in different age group (p value 0.00007 / 0.003 for left mean CIMT and right mean CIMT respectively) while in patients with NAFLD, significant difference was found in mean CIMT of right side and in patients with AFLD, difference was significant on left side in different age groups. These results were consistent with previous study done by Jadhav et al, according to which CIMT value in general population varies between 0.4 to 1 mm. Age is important determinant factor. CIMT increases 0.01 to 0.33 mm/year. This progression of thickness is 0.03-0.06 mm/year in patients with CAD.

When mean CIMT of both side were compared in a particular age group between NAFLD, AFLD and controls, significant difference was found in mean CIMT in age group from 40-49 years in our study (p value 0.03 for right mean CIMT and 0.002 for left mean CIMT). Mean CIMT of both sides was higher in AFLD group in comparison to NAFLD and controls.

Targheret al showed a correlation between higher CIMT and ultrasonographic grade of liver disease. In our study the mean CIMT (left) in ultrasonographic grades I, II and III in NAFLD group was 0.60±1.3mm, 0.82±0.16mm, 0.95±0.07 mm respectively and is statistically significant (p value 0.0001). Similarly, in AFLD group the mean CIMT (left) in ultrasonographic grades I, II and III was 0.66±0.07mm, 0.73±0.12mm and 0.90±0.14mm respectively and was statistically significant (p value 0.006). This showed that as grades of fatty liver increases, mean CIMT also increases. Similar relation is true for mean CIMT of right side.

In our study, mean CIMT when compared in NAFLD patients with and without risk factors for atherosclerosis was found to be significantly more on right side (p value 0.03) in patients whom risk factors for atherosclerosis were present. Similarly, in AFLD patients mean CIMT (It) was significantly more when risk factors were present. In both the groups, risk factors for atherosclerosis were associated with increased CIMT.

Control group consist of cases without fatty liver and were further divided into cases with and without risk factors for atherosclerosis. So, on comparison of mean CIMT between NAFLD patients without risk factors and patients in control group without risk factors, mean CIMT of both side in a NAFLD patients was found to be significantly more (p value 0.004) indicating that NAFLD per se was associated with increased CIMT.

Similarly, on comparison of mean CIMT of both sides between AFLD and control group without risk factors, mean CIMT of both side in AFLD group was found to be significantly more (p value 0.02, 0.04 for left and right respectively) indicating that AFLD per se was associated with increased CIMT. In our study mean CIMT increased both in patients of NAFLD and AFLD. The results obtained is justified by a study done by Kim JH et al, in this study patients were divided into group A (no fatty liver without alcohol history), group B (NAFLD), group C (AFLD) and group D (no fatty liver with alcohol history). The control IMT results were compared across all groups. The mean CIMT was 0.55±0.1 mm for group A, 0.6±0.1 mm for group B, 0.59±0.1mm for group C, and 0.54±0.1mm for group D. There was significant difference between group A and B, group A and C and group C and D (p value >0.05), but there was no difference between group B and C (p value 0.736) and proved that "patients with fatty liver have increase CIMT both in NAFLD and AFLD patients".

A population based study also showed a higher prevalence of carotid atherosclerosis plaques in NAFLD patients. In a study it was reported that the mean CIMT above 0.8 mm was associated with a higher prevalence of CAD. Considering mean CIMT > 0.8 mm to be significantly associated with CAD, 43.47% NAFLD patients, 28.57% AFLD patients and 28.75% controls had CIMT >0.8 mm in our study.

Brea A et al in their study concluded that patients with NAFLD had advanced carotid atherosclerosis and NAFLD is an independent predictor of an increase in CIMT. Similarly Assay N et al, concluded that patients with NAFLD, even without metabolic syndrome were at higher risk for atherosclerosis and assessment of NAFLD may be helpful for cardiovascular risk stratification.
Silvia Soookoion, Carlos J. Pirola, concluded that CIMT is strongly associated with NAFLD, showed that patients with hepatic steatosis had an increase of 13% of IMT in comparison to individuals without fatty liver. Result of our study, that NAFLD per se is a risk factor for atherosclerosis was supported by above mentioned studies.

CONCLUSION

CIMT was found to increase with advancing age in all three groups i.e. NAFLD, AFLD and control group. CIMT was more in patients of fatty liver disease (both NAFLD and AFLD) having risk factor for atherosclerosis as compared to those without risk factors and control group. CIMT was found to increase with increasing grades of fatty liver diagnosed by ultrasonography.

All NAFLD and AFLD patients should be investigated for carotid atherosclerosis, as its early detection and management may be helpful in limiting the inherent complications of atherosclerosis.

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