Abstract

**Aim:** To examine the relationship between carotid intima media thickness (CIMT) and silent cerebral infarction (SCI) in order to determine whether CIMT is a predictor of SCI in patients of type 2 diabetic nephropathy.

**Methods:** A total of 80 patients of type 2 diabetic nephropathy were selected on the basis of fasting and 2-hour post-prandial blood sugar, 24 hrs albumin estimation in urine, urea and creatinine in the serum. The selected candidates underwent MRI brain and carotid B mode ultrasonography to find out the event of SCI and to evaluate the CIMT respectively.

**Results:** The SCI was found in 30 (37.5%) patients with type 2 diabetic nephropathy. The mean age, BMI, blood pressure (BP), macroalbuminuria, lipid, low GFR, duration of diabetes and CIMT were significantly higher in the subject with SCI than in those without it. Multiple logistic analyses indicated that age, BP, and CIMT were found to be significant and independent risk factors of SCI in type 2 diabetic nephropathy subjects.

**Conclusion:** CIMT is a surrogate and reliable predictor of higher risk of SCI among type 2 diabetic nephropathy patients.

Introduction

Silent cerebral infarctions (SCIs), also termed covert infarcts or simply MRI infarcts, are parenchymal lesions that have the MRI characteristics of previous infarcts but have not been associated in that individual with clinical signs or symptoms corresponding to a stroke. It is called “silent” because there may be no observable symptoms. It is considered a precursor of symptomatic stroke and progressive brain damage that may be associated with vascular dementia and decreased cognitive functions. Recent studies have shown the prevalence of SCI on MRI scans to range from 5.84% to 28%.

Silent cerebral infarcts have come up as the complications which might involve the microvascular complications – and with the coexistence of diabetes the impending risk of these complications as well as the silent cerebral infarcts increase manifold. Recent research has shown an increased association between silent cerebral infarct and diabetic nephropathy.

The measurement of intima-media thickness of the common carotid artery (CCA-IMT) by ultrasonography has been recognised as a powerful tool to identify subclinical atherosclerosis. Increased CCA-IMT is reported to be associated with stroke in subjects including those with diabetes.

Unfortunately, despite a strong theoretical and logical background, there is limited work on this issue. Clinical research in varied environments will strengthen the
relationship between silent cerebral infarct, carotid intima media thickness and diabetic nephropathy, and hence provide an opportunity to understand this relationship in a better way. If such relationship could be established, then silent infarcts which are otherwise asymptomatic could be detected earlier among the patients exposed to these risks. As silent infarcts often come without a prior symptomatic manifestation and result in a major physical and psychosocial loss, it is always of interest to weigh the risk of silent infarcts among high risk patients.

The present study is an attempt to explore the issue in order to find out an association between carotid intima media thickness and silent cerebral infarction in patients of type 2 diabetic nephropathy.

Material and Methods

The study included 80 patients of type 2 diabetes mellitus with nephropathy attending the Medicine OPD of Era’s Lucknow Medical College, Lucknow from January 2010 to June 2011. All the cases were evaluated as inpatients in one unit of the Department of Medicine. The cases were selected after satisfying all the exclusion/inclusion criteria.

Inclusion criteria

1. Type 2 diabetes mellitus with nephropathy irrespective of other risk factors (hypertension, hyperlipidaemia, obesity, etc.)
2. Age between 30 and 70 years
3. Informed consent to participate.

Exclusion Criteria

Patients having clinical evidence of cerebrovascular stroke (focal neurological deficit) or past history of cerebrovascular stroke, transient ischaemic attack (TIA), other neurological disorders, valvular heart disease, atrial fibrillation, or myocardial infarction.

The study protocol was duly approved by the ethical committee of our institution.

A detailed history and physical examination were recorded, with emphasis on age, gender, duration of diabetes, brachial blood pressure, height, weight, BMI, waist-hip ratio, and fundoscopy. Patients were subjected to the following investigations:

1. Complete blood count
2. FBS and PPBS
3. Glycosylated HbA₁c using the glycated haemoglobin kit in whole blood at 415 nm
4. Urine R/M and 24-hr urine albumin estimation by an immunoturbidimetric assay
5. ECG
6. Lipid profile
7. Blood urea and serum creatinine
8. Serum sodium and potassium
9. MRI brain (to detect SCI)
10. Ultrasonographic scanning of the carotid arteries (to determine CIMT)

Ultrasonographic scanning of the carotid arteries was performed by the LOGIQ 5 GE Medical Equipments, Mumbai. It was measured by a linear probe at 9-11 MHz frequency on a B-mode ultrasound and Color Doppler. IMT was measured in the common carotid artery at 1 or 2 cms from the flow divider. The scanning session lasted for an average of 30 minutes. The IMT, as defined by Pignoli et al11 was measured as the distance from the leading edge of the first echogenic line to the leading edge of the second echogenic line. The first line represents the lumen intimal interface and the second line is produced by the collagen containing upper layer of the tunica adventitia. The image was focussed on the posterior far wall and four images of the left and right common carotid arteries each were recorded at least 15 mm proximal to the bifurcation. The CIMT was calculated as the mean of eight measurements. The presence of plaques and degree of stenosis was noted and analysed. Plaque was defined as a localised lesion of thickness ≥ 1.0 mm; stenosis was defined as > 50% occlusion with systolic frequency peak ≥ 4.0 KHz and spectral broadening. Any focal thickening of intima media ≥ 2.0 mm was excluded. Normal CIMT values 0.6-0.86 mm.12

The patients were divided into four equal quartiles I, II, III and IV of 20 subjects each based on increasing thickness of carotid intima media.

MRI was performed by 0.4 Tesla Hitachi Aperto Machine. SCI lesions were defined as high intensity areas identified on T2-weighted image, coinciding with low intensity areas on T1-weighted image, which was ≥ 3 mm and < 20 mm in diameter. The diagnosis was made when a lesion was surrounded by a hyperintense gliotic rim on fluid attenuated inversion recovery images to exclude dilated perivascular space.

The patients were divided into two groups based on the presence of lesions of SCI, Group I (Non-infarct) and Group II (Infarct).

The statistical analysis was done using SPSS (statistical package for social sciences) version 15.0 statistical analysis software. The values were represented in number (%) and mean ± SD. The data was analysed using chi square test to study the association between various factors. Level of significance was estimated with 95% confidence intervals and p value.
Results

A total of 80 patients of type II diabetic nephropathy were enrolled in the study as per the inclusion/exclusion criteria detailed above. The incidence of silent cerebral infarct in patients of type II diabetic nephropathy in present study was 30/80 (37.5%). On the basis of MRI findings the patients were divided into two groups for evaluation of various risk factors for SCI as follows:

Group I (n=50) : Patients of type II diabetic nephropathy with no evidence of silent cerebral infarct

Group II (n=30) : Patients of type II diabetic nephropathy with an evidence of silent cerebral infarct

It was observed that majority of patients in Group II belonged to age group of 61-70 years whereas in Group I all the patients were within 31 to 60 years of age group. Mean age of subjects in Group I was 50.06 ± 5.78 years whereas the same in Group II was 62.03 ± 4.69 years. Statistically, there was a significant difference in age of patients in two groups (p < 0.001).

There were 31(38.75%) females and 49(61.25%) males in the study. Of 50 patients in Group I, 20(40%) were females and 30(60%) were males. In Group II, there were 11(36.7%) females and 19(63.3%) males. Genderwise no statistically significant difference was observed (p = 0.767).

The mean weight of Group I patients was 58.68 kg while that of Group II patients was 63.07 kg. 159.38 cm was the mean height of Group I patients and 159.63cm of Group II patients. The mean BMI of Group I patients was 23.11 whereas of Group II patients was 24.80. All the anthropometric measurements studied here i.e. weight, height and BMI showed no statistically significant difference between Group I and II (p > 0.05).

Overall 58.75% patients had normal BMI (< 25), 35.0% were overweight (BMI 25 – 29.9) and 6.25% patients were obese (BMI > 30). The majority (76.6%) of normal weighted subjects belonged to Group I. In overweight and obese category majority of subjects belonged to Group II. This difference was statistically significant (p = 0.008).

15 (18.75%) patients had hypertension. In Group I, 5 (10%) patients were hypertensive while in Group II, 10 (33.3%) patients were hypertensive. Thus, majority of subjects with history of hypertension (66.6%) belonged to Group II and majority of non-hypertensive subjects (69.2%) belonged to Group I. This distribution was statistically significant (p = 0.01).

Serum cholesterol levels in 59 (73.8%) patients were within desirable range (≤ 200 mg/dl), in 18 (22.5%) patients were in borderline high range (200-239 mg/dl), while in 3 (3.8%) patients were in high range (> 240 mg/dl). As compared to Group II (40%), significantly higher proportion of subjects in Group I (94%) had serum cholesterol levels within desirable range. Exactly half (50%) the subjects in Group II while only 3 (6%) in group I had borderline high serum cholesterol levels. None of the patients in Group I, but 3 (10%) subjects in Group II had high serum cholesterol levels. Statistically, there was a significant difference between two groups (p < 0.001).

55 (68.8%) patients had low triglyceride levels (< 150 mg/dl), 24 (30%) had levels within normal range (150-260 mg/dl) while only 1 (1.3%) patient had significantly high level (> 260 mg/dl). Proportion of subjects with low triglyceride levels was significantly higher among patients of Group I (84.0%) as compared to that in Group II (43.3%) whereas the proportion of subjects with normal triglyceride levels was significantly higher in Group II (56.7%) as compared to Group I (14.0%). There was 1 (2%) patient in Group I with serum triglyceride levels in high range as against none in Group II. Statistically, there was a significant difference between two groups (p < 0.001).

66 (82.5%) patients had normal serum HDL levels (40-60 mg/dl) and 7 (8.8%) patients had low (< 40 mg/dl) and high (> 60 mg/dl) levels each. All the subjects in Group II had serum HDL levels within normal limits. However, the proportion of subjects with low as well as high HDL levels (14% each) was significantly higher among group I patients (p < 0.001).

52 (65%) patients had serum LDL levels within the normal range (< 100 mg/dl), 14 (17.5%) patients had near optimal levels (100-129 mg/dl), 12 (15%) patients had borderline high levels (130-159 mg/dl) and 2 (2.5%) patients had high (≥ 160 mg/dl) serum LDL levels. None of the subjects in Group I had borderline high or high LDL levels whereas in Group II, a total of 14 (46.7%) patients had LDL levels in borderline high and high range. Statistically, there was a significant difference between the two groups (p < 0.001).

A total of 59 (73.8%) subjects had 24-hr urine albumin levels ≤ 300 mg (microalbuminuria) whereas a total of 21 (26.3%) subjects had 24-hr urine albumin levels > 300 mg (macroalbuminuria). In Group I 46 (92.0%) subjects had microalbuminuria as compared to 13 (43.3%) in Group II. Only 4 (8.0%) subjects from Group I had macroalbuminuria as compared to 17 (56.7%) from Group II. Number of subjects with microalbuminuria was significantly higher in Group I (p < 0.001) as compared to those in Group II while a larger proportion of subjects in Group II had macroalbuminuria as compared to Group II.

Overall 29 (36.3%) patients, 18 (36%) in Group I and 11 (36.7%) in Group II had GFR values between...
Association between CIMT and SCI

To study the association between CIMT and SCI, all the 80 patients were divided into four equal quartiles I, II, III and IV of 20 subjects each based on increasing CIMT. In quartile I, the CIMT of the patients was in the range 0.5 to 0.6 mm; in quartile II, the CIMT was in the range of 0.6 to 0.7 mm; in quartile III CIMT of the patients was in the range of 0.7 to 0.9; whereas in quartile IV CIMT of the patients was ≥ 0.9.

A positive association between increase in CIMT and incidence of silent cerebral infarct was found (p < 0.001). It was observed that among patients in lowest quartile of CIMT the incidence of SCI was 0% whereas in highest quartile it was 100%. The incidence of SCI in the II and III quartile was 5% (19) and 45% (9) respectively. The relative risk (RR) of SCI in I and II quartile was 0.04 whereas the RR of SCI in quartile III and IV was 4.39.

All the 20 patients (100%) in the first quartile had diabetes since less than 5 years. All 20 patients (100%) belonging to the fourth quartile had diabetes of longer (> 5 yrs) duration. Only 1 patient (5%) in the second quartile and 12 patients (60%) in the third quartile had diabetes for more than 5 years. A significant association between duration of diabetes and increasing CIMT was observed. It was observed that with increasing duration of diabetes the proportion of subjects in higher quartiles of CIMT increased (p < 0.001).

Discussion

A total of 80 patients of type II diabetic nephropathy were enrolled in the present study. Out of these, 30 patients were found to have SCI on MRI brain. Thus the incidence of silent cerebral infarct among patients of type II diabetic nephropathy was 37.5% in our study. In a previous study, Uzu et al.8 had reported SCI in 177 out of 608 (29.1%) patients with diabetes. Nomura et al. (2010)13 found an incidence of SCI in 131 out of 217 (60.4%) patients with diabetes, which was much higher as compared to the present study. The reason for higher incidence might be higher mean age of subjects enrolled in the study (67.1 ± 8.6 years) as compared to mean age of 54.55 ± 7.93 years in the present study. Thus, the incidence of SCI in present study is similar to that noted in the study of Uzu et al. This is a very high incidence and depicts a high risk of cerebral stroke which may be life-threatening or might cause serious physical and mental impairment. This high incidence indicates the need to take special care of type II diabetic nephropathy patients in view of impending threat of silent cerebral infarct.

As far as the prevalence of SCI in different age groups among patients of type II diabetic nephropathy is concerned, it was maximum in age group of 61-70 years (n=17/17; 100%) and minimum in age group of 31-40 years (n = 0/3; 0%). The mean age of subjects with SCI was found to be 62.03 ± 4.69 years. In the study of Uzu et al.8 the mean age of patients was 63.3 ± 7.8 years. However, in study of Nomura et al.13 The mean age of patients with SCI were observed to be 69.3 ± 6.9 years. It would be pertinent to mention here that in present study the patients were having type II diabetes with nephropathy whereas the study of Nomura et al.13 had included the patients of diabetes mellitus only. Apart from that the study of Nomura et al was carried out among Japanese patients; Japan is traditionally considered to be having a higher life-expectancy and superior health awareness. Thus the findings of the present study are in accordance with those quoted in literature. This observation indicates the need for periodic screening among the patients of type II diabetic nephropathy above 50 years of age.

Gender wise no significant difference in incidence of SCI was observed in this study. Uzu et al.8 too did not find a significant association between gender and SCI. Nomura et al.13 also did not find a gender associated risk of SCI. Thus our findings are in concordance with that of other studies.

In this study no significant difference between SCI and non-SCI subjects was observed in mean anthropometric parameters i.e. weight, height and BMI. Uzu et al.8 too did not find a significant difference in mean weight, height and BMI of two groups. However, on categorical comparison of BMI, it was observed that significantly higher proportion of subjects in overweight and obese category had SCI. This is another important finding for screening the risk of SCI among type II diabetic nephropathy patients.

In the present study, significantly higher proportion of subjects with hypertension (66.6%) had SCI as
compared to those not having hypertension (30.8%) (p = 0.01). Nomura et al\textsuperscript{13} observed the mean systolic blood pressure to be significantly higher in subjects with SCI than in those without it. Ricci et al\textsuperscript{14} too found a higher incidence of silent cerebral infarction in patients with hypertension. Vermeer et al\textsuperscript{8} have found raised blood pressure to be associated with silent cerebral infarct in the age group 60 - 90 years. Hypertension is a known risk factor for stroke.

In the present study, a significantly higher percentage of subjects with SCI had borderline high to high serum total cholesterol levels (p < 0.001). However, no such association was observed by Uzu et al.\textsuperscript{8} To the best of our knowledge, most of the other studies have not reported any association between serum total cholesterol levels and incidence of SCI. Majority of subjects in our study in non-SCI group had subnormal levels of triglycerides whereas majority in SCI group had triglyceride levels within normal limits thereby showing a significant difference between two groups (p < 0.001). Neither Nomura et al\textsuperscript{13} nor Uzu et al\textsuperscript{8} who carried out extensive study among SCI patients with diabetes reported any association between any of lipid profile level and SCI. Ong et al\textsuperscript{15} too did not find a significant association between SCI and hypertriglyceridaemia. In present study too we did not find an association between hypertriglyceridaemia and SCI, however, categorically majority of subjects in non-SCI group had subnormal values. It would be pertinent to mention here that in general, triglyceride levels show a high variability as reported by Nomura et al\textsuperscript{13} and Uzu et al\textsuperscript{8} who reported high standard deviation values for serum triglyceride levels. The lipid control in diabetics is subjective which is generally dependent on the individual’s lifestyle. As high variability is expected, it might be a chance occurrence that the subjects in SCI group had higher proportion of subjects with higher serum triglyceride levels. In the present study, for serum HDL levels a statistically significant difference (p < 0.001) between two groups was observed. At the one hand all the subjects in SCI group had normal HDL levels, on the other hand 7 out of 50 (14%) subjects in non-SCI group had HDL levels below normal and another 7 out of 50 (14%) subjects had HDL levels above normal. In this situation it is difficult to find out the substantiability of any association between the two. Nomura et al\textsuperscript{13} have not found any association between serum HDL levels and SCI. Uzu et al\textsuperscript{8} also ruled out such an association. In this study, for serum LDL levels, statistically highly significant (p < 0.001) proportion of subjects in SCI group had borderline high to high levels as compared to those in non-SCI group. However, Nomura et al\textsuperscript{13} have not found any association between Serum LDL levels and SCI.

Most other studies performed on Western population or population from the developed world have not reported an association between lipid parameters and incidence of SCI. The findings in our study which includes Indian subjects show a significant association between SCI and lipid parameters. The reason for this association might be the higher lipid levels in the general Indian population, which needs further substantiation.

In the present study, a statistically highly significant difference was observed in urine albumin levels of SCI and non-SCI group (p < 0.001). Out of 21 patients with macroalbuminuria, 17 (81%) had silent cerebral infarct. None of the subjects in present study had urinary albumin levels in the nephrotic range (> 3500 mg/24 hr). Similar results have been reported by Uzu et al\textsuperscript{8} who found a significantly higher proportion of subjects with albuminuria among SCI subjects. Nakamura et al\textsuperscript{17} in a drug trial reported that incidence of SCI in type 2 diabetes patients with albuminuria could be reduced by dilazep dihydrochloride. As all the patients were under treatment for a prolonged duration hence it is difficult to comment whether any treatment has an altering effect on the incidence of SCI among patients in different categories of urinary albumin levels.

On seeking the association between glomerular filtration rate and incidence of SCI, it was observed that SCI patients had significantly higher percentage of subjects with lower values of GFR than non-SCI patients (p < 0.001). Uzu et al\textsuperscript{8} also had similar observations. Kobayashi et al\textsuperscript{18} too found a similar relationship between SCI and estimated glomerular filtration rates.

In present study, a significant association between incidence of SCI and duration of diabetes was observed. Median duration for development of angiopathy among diabetic patients has been reported to be around 10 years,\textsuperscript{19} thus with increasing duration of diabetes the incidence of microvascular complications and SCI increases. In present study we tried to explore the relationship on a five-yearly basis, as target end organ damage is expected within 11 to 15 years. We observed that all (n=10/10; 100%) patients with diabetes for > 11 years all the patients had evidence of SCI. This is once again an interesting finding from the point of view of screening. The proportional risk between ≤ 5 years to that of > 5 years was 41:1.

Recently, CIMT has emerged as a predictive marker for assessment of risk of SCI.\textsuperscript{13} It is an easy, non invasive and cheap method to assess atherosclerosis and its impact on different associated cardiovascular co-morbidities. As limited literature related with risk assessment of SCI among diabetic nephropathy patients is available, we relied on a descriptive
analytical method by exploring the incidence of SCI among different quartiles of CIMT in increasing order. The CIMT was measured for both left and right sides, however the higher of the two values was taken into account. It was observed that except for one, all cases of SCI were in third and fourth quartiles, whereas the proportion of subjects in non-SCI group was higher in first and second quartiles of CIMT, thus showing a significant impact of increasing CIMT on the incidence of SCI. In higher quartiles (III, IV) of CIMT the relative risk of SCI was much higher as compared to lower quartiles (I, II) (RR: 4.39 vs. 0.04). Nomura et al. (2010) also observed the mean CIMT of patients with SCI to be significantly higher as compared to non-SCI patients of type 2 diabetes. Similar observations have been made by Das et al. who observed Odds ratio of SCI among patients with increased CIMT to be 1.65. However, Inoue et al. found no significant independent role of CIMT in prediction of SCI, instead they associated coexistence of increased CIMT with plaque as a higher risk for SCI. Increased CIMT is reported to be associated with stroke in subjects including those with diabetes. Eguchi et al. too supported these findings while Johnsen et al. was of the view that CIMT was associated with hypertensive vascular disease. In present study, we too found similar relationship. Kozera et al. indicated that presence of diabetic nephropathy, but not IMT can be regarded as an indicator of cerebral microangiopathy severity in patients with type 2 diabetes. However, in present study that included only the patients of type II diabetic nephropathy, the higher incidence of SCI among those in higher quartiles of CIMT proves existence of such relationship with CIMT too, though the cut-off values for diabetic alone and the patients with diabetic nephropathy might vary. The present work does not propose any particular cut-off value as it would be inappropriate to reach on to a conclusive cut-off marker without ascertaining the variable profile of individuals including the duration of disease, associated illnesses, level of complications, age, gender, lipid profile and level of glycaemic control. An interesting finding during the course of study was that a total of 11 patients in third quartile of CIMT did not have infarct, among these 7 (63.6%) had HDL levels above 58 mg/dl. This is an interesting finding which also indicates that CIMT as a single predictor cannot be and should not be proposed as a determinant of SCI without taking into account other variables.

On seeking the relationship between duration of diabetes and CIMT, it was observed that the patients with higher CIMT values were mostly those with higher duration of diabetes. We also found a significant association between SCI and duration of diabetes. Thus a common association of CIMT and SCI with duration of diabetes could be observed. Similarly, on seeking the association between infarction, urinary albumin levels and CIMT, it was observed that majority of patients with higher quartiles of CIMT had SCI and/or macroalbuminuria. This is another finding that needs further exploration.

**Conclusion**

CIMT is independently associated with type II diabetic nephropathy. CIMT is a surrogate and reliable predictor of higher risk of SCI in type II diabetic nephropathy patients implying that patients with high CIMT should be screened for SCI, which is a risk factor for stroke and dementia. More intensive preventive management, including active detection of SCI and strict treatment of multiple cardiovascular risk factors, especially hypertension is indicated.

**References**


