Detection of Subclinical Atherosclerosis in Peripheral Arterial Beds With B-Mode Ultrasound

A Proposal for Guiding the Decision for Medical Intervention and an Artifact-Corrected Volumetric Scoring Index

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ABSTRACT

Objectives: To assess subclinical atherosclerotic cardiovascular disease (ASCVD) using B-mode ultrasound, with special emphasis on the incremental value of performing imaging in multiple peripheral arteries, and to compare imaging findings with traditional risk factors for medical intervention eligibility.

Methods: Data from 2 asymptomatic cohorts from India with unknown ASCVD risk factors were compared to 2 cohorts from North America with known ASCVD risk factors. Carotid and iliofemoral arteries of the Indian cohorts were examined with automated ultrasound in a high-pace environment by non-experts. A simplified metric of atherosclerotic disease burden (FUster-Narula or FUN Score) was developed from 3D imaging data by summing intima-media volume (IMV) over 5-cm arterial segments. Effectiveness of ASCVD prevention guidelines to direct therapy was compared to results from direct imaging.

Results: Of the 941 (mean age 44.27 ± 13.76 years, 34% female) enrollees from India, 224 (24%) demonstrated plaques in at least 1 of the 4 arterial sites examined; 107 (11%) had plaques in only the carotids, 70 (7%) in both the carotids and iliofemoral arteries, and 47 (5%) had plaques in only the iliofemoral arteries. Older age and male sex were associated with the presence of plaque, but association with systolic blood pressure was not observed.
Data from 2 North American clinics (n = 481, mean age 59.68 ± 11.95 years, 39% female) showed that 203 subjects (42%) had carotid plaque; 82% of whom would not have qualified for lipid-lowering therapy under the Adult Treatment Panel (ATP) III Guidelines. Using the recently published ATP IV Guidelines, 33% of the individuals with carotid plaque would also have failed to qualify for treatment.

Conclusions: B-mode ultrasound examination of bilateral iliofemoral arteries provided an incremental yield in identifying subclinical atherosclerotic disease compared to carotid evaluation alone. Ultrasound examination allowed improved identification of individuals who could be targeted for prophylactic medical intervention compared to ATP III and ATP IV Guidelines.

Prognostic stratification strategies based on traditional risk factors have been effective in reducing morbidity and mortality from atherothrombotic events in developed health care systems over the last 30 to 50 years [1,2]. The rapid rise in obesity, metabolic syndrome, diabetes mellitus, prevalence of hypertension, gross changes in lifestyle and behavioral modification [3], as well as the widespread use of lipid-lowering medications has led to some disillusionment regarding the universal applicability of traditional risk factors as the sole predictors of events. Atherosclerotic cardiovascular disease (ASCVD) risk prediction algorithms are not very well understood or followed in developing countries [4]. Furthermore, traditional ASCVD risk factors demonstrate a modest specificity and a variable relationship to the development of atherosclerotic disease; subjects exposed to atherogenic risk factors do not always go on to develop ASCVD events and the disease may also develop in the absence of overtly abnormal risk factors. Data from the Framingham Heart Study with a 26-year follow-up revealed that there was a significant overlap in risk factors between those who suffered a myocardial infarction and those who did not; for example, cholesterol levels were the same in 80% of the patients who did and did not experience a myocardial infarction [5]. In a more recent retrospective

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study of 232,000 patients admitted to hospitals with coronary artery disease, low-density lipoprotein levels were <130 mg/dl in >70% of patients and high-density lipoprotein ≥40 mg/dl in almost one-half of the patients [6].

Atherosclerosis is a systemic process. Numerous clinical, epidemiological, and pathological studies have shown that atherosclerosis develops as a diffuse process in the central and peripheral arterial system [7–10] and that chronic advanced atherosclerosis can exist with minimal or no symptoms. Mortality rates from ASCVD-related complications among men and women of working age in emerging economies such as India are significantly higher than they are in industrialized countries [11–14]. To reduce the societal impact of this epidemic, the decision for initiating preventative medical intervention based solely on traditional risk factor assessment needs to be re-evaluated. It is conceivable that prophylactic medical intervention might be timely (if not already late) in asymptomatic subjects who exhibit direct evidence of atherosclerotic lesions in easily accessible peripheral arterial beds, irrespective of the presence of overt risk factors [15–19]. This is because lesions in the peripheral arteries not only might lead to major adverse events but also because peripheral arterial involvement often precedes coronary artery involvement in the progression of atherosclerosis [20,21]. The strategy to treat individuals with direct evidence of subclinical atherosclerosis might be particularly applicable where traditional risk factor–based algorithms are not well established or readily available, as is the case in most low- and middle-income countries.

In view of the latest American College of Cardiology/American Heart Association (ACC/AHA) and European Society of Cardiology [22] recommendations for prescribing lipid-lowering agents to individuals with pre-existing evidence of atherosclerotic disease, the use of imaging tests that demonstrate direct evidence of atherosclerosis might provide an ideal strategy to guide the decision for initiating medical intervention. Compared with other imaging modalities, ultrasound provides a safe, portable, and low-cost solution that is most amenable to mass adoption in the primary care setting. In a large European study involving over 10,000 relatively young asymptomatic subjects with a 10-year follow-up, ultrasound examination of 4 peripheral arterial beds correctly identified 99% of future ASCVD events [23]. Doppler ultrasound–based flow studies are routinely used in clinical practice for the assessment of carotid stenosis in symptomatic patients to identify those who would benefit from surgical intervention. With recent improvements in image quality and built-in automation, perhaps B-mode ultrasound can similarly be deployed to assess subclinical atherosclerosis in the asymptomatic population to identify subjects who would benefit from prophylactic medical intervention.

In an earlier study, clinical utility of B-mode ultrasound examination of the extracranial carotid system in an asymptomatic population in northern India was assessed [4]. The present study expands on this work by incorporating examination of the iliofemoral arteries. The main aim of this study was to determine the prevalence of atherosclerotic disease in relatively young asymptomatic individuals in 2 underserved communities in India using B-mode ultrasound examination of 4 peripheral arterial beds where ASCVD risk factor information was not available. A quantitative method was developed to summarize ultrasound examination findings and create a yardstick to help generate uniform data going forward. We also sought to compare prevalence of subclinical atherosclerotic disease in a developed health care system and to assess the value of direct imaging, compared with standard ASCVD risk factor algorithms, to initiate preventative medical treatment.

METHODS

Data collection

Ultrasound imaging data from 4 arterial beds were collected from 2 different communities in northern India and were compared with reference data collected from 2 clinics in North America.

Free health-check camps staffed by trained radiology residents in the semiurban town of Sirsa (Haryana) and the urban city of Jaipur (Rajasthan) enrolled 941 asymptomatic volunteers (mean age 44.27 ± 13.76 years, 34% female) over 8 consecutive days. The community in Sirsa was expected to be relatively free of traditional ASCVD risk factors because the study enrollees were devout followers of a local spiritual leader—Sant Gurmeet Ram Rahim Singh Maharaj—who discourages consumption of tobacco products, meat, eggs, fish, or alcohol. Physical activity and meditation, on the other hand, are strongly encouraged. None of the participants from Sirsa was a current smoker, though some had smoking histories. Most were laborers tilling fields or involved in similar physically intense activities. The community from the city of Jaipur, which is probably more representative of the wider population of India, included many current smokers who had a higher risk factor profile and led a more sedentary lifestyle.

Appropriate institutional review board approvals were taken and all participants provided written informed consent. A simple questionnaire was used to collect basic information about age, smoking history, hypertension, diabetes, and family history of cardiovascular disease. Female enrollees were specifically asked whether they were pregnant. If so, they were informed of their ineligibility to participate in the ultrasound examination in compliance with local prenatal imaging regulations. Resting blood pressure was measured to identify any undetected hypertension and history of current medications was noted. Blood samples were not drawn.

Reference clinical data

ASCVD risk factors and carotid ultrasound examination data were obtained from 2 North American primary care clinics in anonymized fashion to serve as a reference to allow comparison with data collected in India. One clinic was located in Toronto, Ontario, Canada, and the other in

Richmond, Texas, USA. Comprehensive risk factor profiles were gathered from self-referred asymptomatic patients as part of a preventative annual physical examination. In addition, a bilateral B-mode ultrasound examination of the carotids was administered. Sample clinical data from 481 patients (mean age 59.68 ± 11.95 years, 39% female) were gathered over an 8-week period. Most of the participants in these 2 urban centers were office workers in mid- to high-income brackets; some were current smokers, and few engaged in regular physical activity.

Carotid and iliofemoral artery examination

Eight radiology residents received 2 hours of training on the dedicated ultrasound equipment for conducting the ultrasound examination at 2 health camps organized in India. In the 2 North American clinics, the ultrasound examination was administered by trained vascular sonographers.

Bilateral B-mode ultrasound examination was performed to detect plaque in the extracranial carotid system and measure intima-media thickness (IMT) in a 1-cm segment of the far wall of the common carotid, 1 cm proximal to the flow divider. This exam was repeated in the iliofemoral region to include the common femoral artery and its bifurcation. Participants were screened in the supine position. For carotids, the neck was oriented 45° using a custom pillow, and B-mode ultrasound images were captured with a 9-MHz linear array transducer in accordance with the clinical protocol recommended by the American Society of Echocardiography consensus statement [17]. Presence or absence of plaque in the carotids and iliofemoral arteries was first determined by acquiring images in the short-axis (transversal) view and then reconfirmed in the long-axis (longitudinal) view. If a plaque was found, the ultrasound image was marked up with the help of on-board tools as illustrated in Figure 1. Plaque was defined as any focal wall thickening >1.5 mm protruding into the lumen of the scanned vessel [24]. In the neck, the transducer was manually moved in the cranial direction from the proximal common carotid artery (clavicle) to the highest segment above the carotid bifurcation that was accessible. The iliofemoral arteries were examined from just below the inguinal ligament with little compression maneuver to compress the adjacent common femoral vein. The transducer was moved downward from the common femoral artery to approximately 2 cm below its bifurcation. It should be noted that femoral ultrasound examination was not administered to the 2 cohorts from North America. Therefore, far-wall common femoral IMT data were excluded from the analysis when data from the 2 Indian cohorts were compared with data from North American cohorts.

The ultrasound system used incorporates a sensor in the transducer that tracks the scanning angle in real time and records the transducer angle position at which the IMT is taken. Although the user is able to make multiple angle measurements at predefined locations (anterior/mid/posterior), to save time, a single-angle measurement was taken that showed the clearest visualization of IMT and the straightest artery segment at which maximum IMT was obtained. The ultrasound system also tracks the arterial diameter change continuously and determines systolic and diastolic timing based on vessel lumen diameter changes without requiring an external electrocardiogram. Automated calculation of IMT was done over a 1-cm region of interest based on raw radio frequency data in the far wall of the vessel examined. Within the 1-cm region of interest, the system tracks 24 spatial measurements at 200 frames/s for a total of 4,800 measurements every second. Thus, the reported IMT values from the ultrasound system were an average of 24 spatial measurements over a 1-cm region at end diastole (Figure 1B). The built-in automated functions of the ultrasound technology obviated the need for qualified radiologists or cardiologists to separately make IMT measurements offline.

2D, 3D, and IMV

Conventional 2-dimensional (2D) imaging was deemed adequate for the identification of focal lesions. However, as a necessary step toward reducing or potentially even eliminating operator dependency, plaque identification and quantification needed to be automated. The only viable way

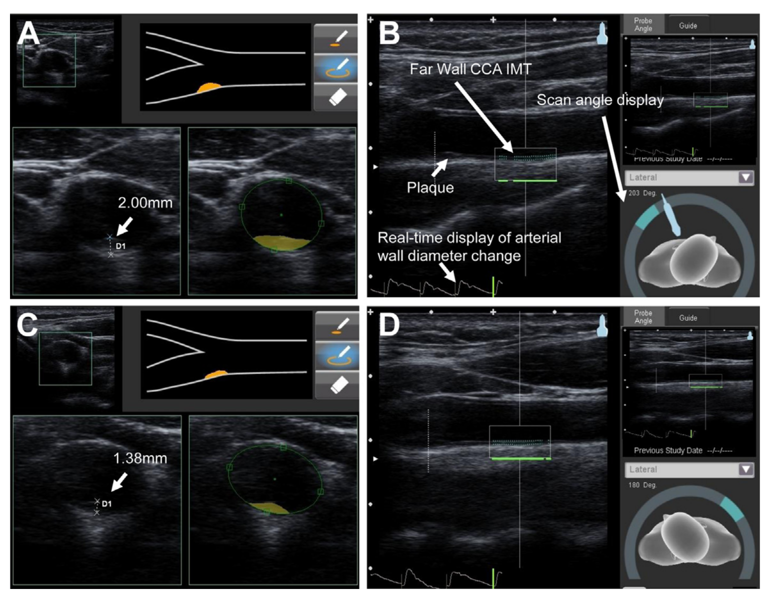

FIGURE 1. A sample screenshot. This is an example of the protocol followed. In this 33-year-old male subject, the left carotid of the subject was scanned in the transversal plane (short-axis view) from the base of the common carotid up through the bifurcation. An atherosclerotic plaque (focally elevated intima-media thickness [IMT] protruding into the lumen by >1.5 mm) was found in the bulb region, and this was marked up (A). The far-wall IMT of the common carotid approximately 1 cm from the flow-divider was then measured and recorded in the longitudinal plane (long-axis view) (B). The examination was repeated on the right carotid, but this time no plaque was identified. The corresponding transversal (C) and longitudinal (D) images are illustrated.

to achieve this goal was to gather 3-dimensional (3D) imaging data for the arterial segment of interest. It should be emphasized that constructing and displaying 3D images of the arteries, though interesting, has limited clinical utility. The more clinically important purpose of gathering 3D data was to automate plaque detection and to quantify it into an actionable index of atherosclerotic disease burden. For illustrative purposes, 4 individuals were selected for calculation of the FUster-Narula (FUN) score (see Technical Considerations). The first step toward constructing the FUN score involved scanning the artery in the short-axis view starting approximately 3 cm proximal to the flow-divider and continuing in a unidirectional manner to beyond 2 cm distal to the flow-divider. Transducer position was tracked optically in 6 dimensions. Artery constituent layers were automatically detected and displayed as contours on each cross-sectional imaging frame from which intima-media area was calculated by the system. Using the position-tracking information, contiguous frames were aligned where necessary and intima-media areas over a standardized 5-cm segment of the artery were summed to calculate the intima-media volume (IMV). This process was repeated bilaterally in the carotids and iliofemoral arteries. Summation of the 4 IMV formed the FUN score with additional notation provided to indicate the presence of focal lesions. At the time of data acquisition, not all the steps for automated contour detection, frame orientation, and imaging artefact correction were available on the ultrasound machine, which necessitated some manual adjustments.

TECHNICAL CONSIDERATIONS

Automated vascular ultrasound imaging for mass screening

Vascular ultrasound imaging has been used in large hospitals and academic centers for >40 years. However, its applicability has not been established in the primary care setting even after the recent availability of low-cost portable ultrasound instruments. Fundamental barriers to mass adoption include the need for expertise in acquiring and interpreting ultrasound images. Traditionally, highly trained sonographers have acquired the data, which in turn are interpreted by equally skilled radiologists, vascular medicine experts, or cardiologists. This overhead of expertise is perfectly suited for handling the relatively small number of symptomatic patients but is ill equipped to handle the much larger asymptomatic population who would benefit the most from imaging technology to guide preventative interventions. To help translate ultrasound imaging methods for the assessment of atherosclerotic disease burden in the asymptomatic population from an academic curiosity to mass deployment requires technological improvements toward automated plaque detection and quantification. Some of the advancements made to the imaging equipment (CardioHealth Station, Panasonic Healthcare, Tokyo, Japan) to test the feasibility of mass screening are described in the following sections.

Transducer. Volumetric imaging data were acquired with a conventional linear array transducer equipped with means for tracking free-hand movement in multiple dimensions. The positioning information provided knowledge about imaging slice interval and its orientation. Computer algorithms facilitated alignment of contiguous imaging frames to reduce plaque quantification errors from 3D reconstructed datasets.

Image artifact. Differentiating between pathological findings and image artifact remains a huge challenge. In arterial ultrasound scanning, the transversal view is the preferred method for identifying focal lesions because it is less prone to certain types of imaging artifacts. Nevertheless, refraction and reflection effects can still introduce artifacts, such as the acoustic shadowing from the edges of curved surfaces [25,26], that need to be corrected.

Tissue interfaces. Accurate determination of the location of lumen-intima, intima-media, and media-adventitia boundaries is another technical challenge. A certain amount of signal integrity is lost during the scan conversion process. Better algorithms were developed that analyze higher fidelity raw radio frequency data for more precisely locating various boundaries of the layers of the arterial wall. This process has been fully automated to reduce operator dependency.

A quantitative index of subclinical atherosclerosis:

FUN score. Clinical findings need to be presented in a standardized quantitative index that is easy to understand and actionable. The utility of elevation in IMT in the common carotid as an index of subclinical atherosclerosis has not succeeded as an independent determinant of ASCVD-related events; numerous studies including a meta-analysis involving >45,000 individuals support this connotation [27]. It should be emphasized that data analyses in such studies did not include focal plaque findings, which are often found distal to the flow divider in the bulb or internal carotid, not in the common carotid. Total plaque area has also been proposed as an index [28], but this method was heavily operator- and equipment-dependent due predominantly to transducer orientation angle variability. Measurement error with this method may not be so noticeable when examining relatively older patients who have large plaques, but for a younger population with small focal lesions, the error in the measurement would likely become prohibitively high. In view of these limitations, the FUN score was proposed [29], which is a summation of IMV in 4 peripheral arteries (bilateral carotids and bilateral iliofemoral arteries) over a 5-cm segment that includes 2-cm segment distal and a 3-cm segment proximal to the flow-divider, see Figure 2. Presence of lesions in the common femoral artery is more frequent than in the common carotid, which was the rationale for selecting a standard 3-cm segment proximal to the flow-divider. The 2-cm segment distal to the flow-divider will encompass the bulb and approximately a 1-cm region into the bifurcation, the location that is particularly prone to developing atheroma. The FUN

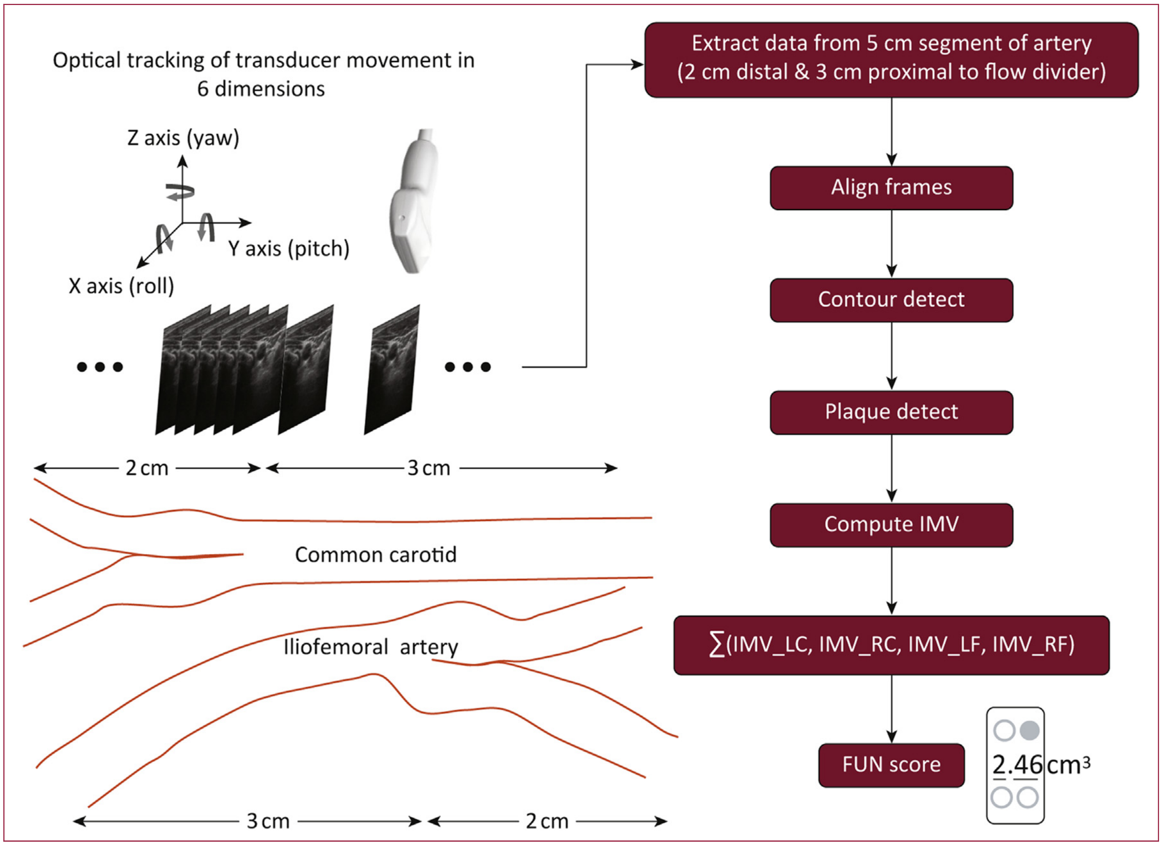

FIGURE 2. Computation of the FUN score. The FUster-Narula (FUN) score is a summation of intima-media volume (IMV) in 4 peripheral arteries (bilateral carotids and bilateral iliofemoral arteries) over a 5-cm segment that includes 2 cm distal to the flow-divider and a 3-cm segment proximal to the flow-divider. The presence of lesions in the common iliofemoral artery is far more frequent than in the common carotid, which is the rationale for selecting a standard 3-cm segment proximal to the flow-divider. The 2-cm segment distal to the flow-divider encompasses the bulb and approximately 1-cm region into the bifurcation, the segment of the artery that is particularly prone to developing atheroma. The FUN score notation includes 4 circles representing each artery examined. A solid circle indicates that a plaque was identified in that artery (any IMT protruding into the lumen >1.5 mm). Please review illustrative examples in Figure 5. LC, left carotid; LF, left femoral; RC, right carotid; RF, right femoral.

score notation includes 4 circles representing each artery examined. A solid circle indicates that ≥1 plaque (focally elevated IMT protruding into the lumen by >1.5 mm) was identified in that artery. The rationale for separately notating plaque findings was to differentiate potentially comparable volumetric FUN scores in those individuals who have systemic elevation in IMV but no plaque from those who may have normal IMV but with focal plaque(s).

RESULTS

Cohorts from medical camps in India

A total of 946 subjects participated in carotid and iliofemoral B-mode ultrasound examinations; 5 were excluded from data analysis (2 did not provide consent for iliofemoral scanning, 1 case had images missing, 2 had very poor image quality). The mean age of the 941 cases analyzed was a relatively young, 44.27 ± 13.76 years (compared with 59.68 ± 11.95 years for the reference cohort from North America). The Indian cohort comprised 624 men (44.59 ± 14.53 years) and 317 women (43.64 ± 12.10 years). Mean age of the constituent cohorts from Sirsa and Jaipur was 43.04 ± 14.20 and 46.88 ± 12.13 years, respectively. Corresponding summary of bilateral far-wall common carotid IMT is illustrated in Figures 3A and 3B. Far-wall common femoral IMT data were excluded from this analysis to facilitate direct comparison with data from North American cohorts where femoral scanning was not performed.

Participants in the semiurban community of Sirsa were known to have undergone aggressive lifestyle modifications. Based on the responses to the questionnaire, 61 male enrollees (15%) identified themselves as former smokers; none were current smokers; 90 (19%) were on medication to control hypertension; and 9 (2%) were on medication for type II diabetes. Sirsa enrollees lead a very active lifestyle. The male urban enrollees from Jaipur led a more sedentary

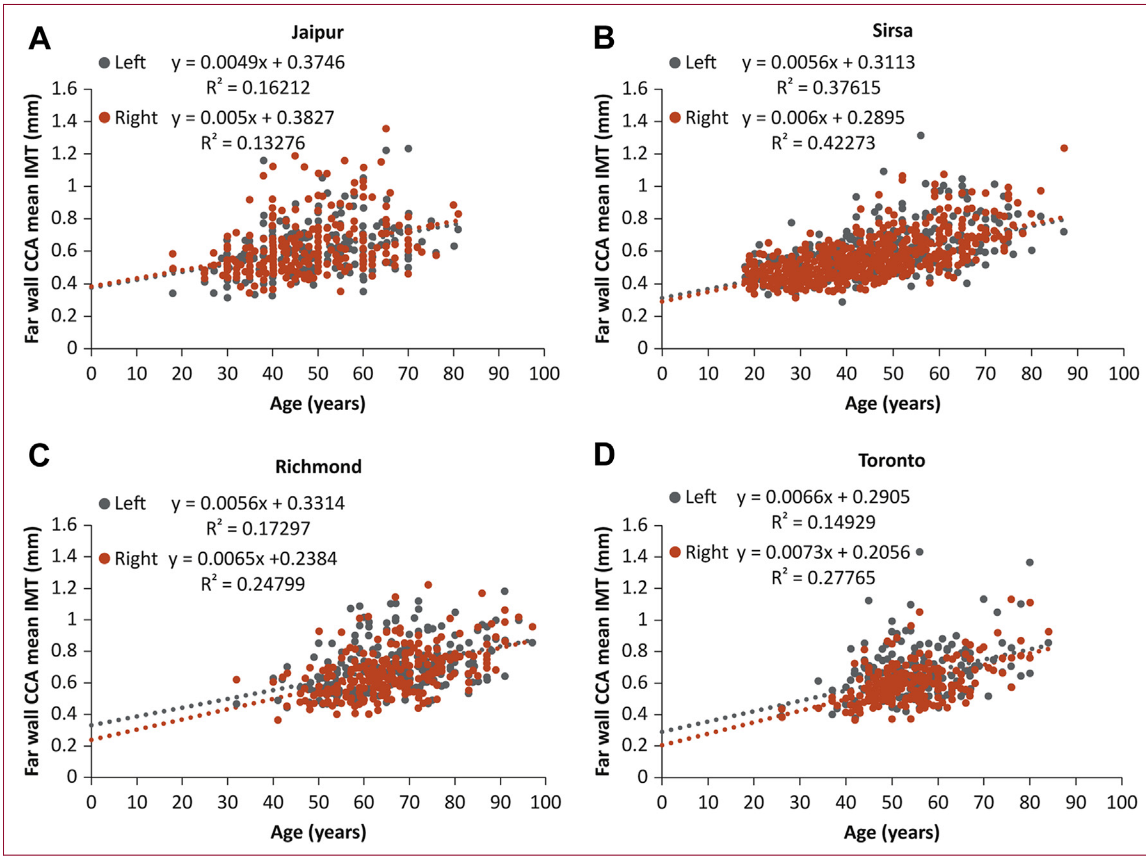

FIGURE 3. Increase in far-wall common carotid IMT with age. Univariate linear regression analysis shows that the slope in the North American cohorts is slightly higher than that for the Indian cohorts. Contributing factors for this observation include lower sample number and higher average age of the North American cohorts. These graphs illustrate why it is so difficult to generalize such results to populations of varying ethnicity, sex, and age as the basis for forming clinical decisions. (A) Jaipur, India. (B) Sirsa, India. (C) Richmond, Texas, USA. (D) Toronto, Ontario, Canada. CCA, carotid coronary artery; IMT, intima-media thickness.

lifestyle; 64 (40%) were current or former smokers; 33 (21%) were on medication to control hypertension; and 10 (6%) were on medication for type II diabetes. None of the women identified themselves as ever having smoked; a finding that is believable given that this is the cultural norm in India.

An analysis of the 941 subjects showed that 224 (23.8%) had atherosclerotic plaques in either the carotid or iliofemoral arteries. Only the carotids were involved in 107 subjects (11.4%) and only the iliofemoral arteries were involved in 47 subjects (5.0%). In 70 subjects (7.6%), plaques were found both in the carotids and iliofemoral arteries. A summary of plaque findings in different arterial sites is presented in Figure 4. Prevalence of plaque in the male cohort was higher (n = 191, 30%) than in the female cohort (n = 33, 10%, $p < 0.0001$). Older age and male sex was significantly associated with the presence of plaque both in urban (58 ± 12 years, $p < 0.0001$) and semiurban populations (57 ± 12 years, $p < 0.0001$). However, systolic blood pressure was found to be the same in those with and without plaques (140 ± 30 vs. 135 ± 18, p = NS) in both populations. Plaque prevalence in male subjects in the semiurban population of Sirsa was significantly lower than in the male cohort of urban Jaipur (27% vs. 41%, respectively, $p < 0.001$). Male smokers (former and current) from Sirsa and Jaipur had significantly higher prevalence of plaque (n = 61, 49%) than did nonsmokers (n = 130, 26%, $p < 0.0001$). For illustrative purposes, the FUN score was calculated step-wise and is presented for 4 asymptomatic individuals, see Figure 5.

Inter- and intraobserver variability in automated IMT measurement was not undertaken because the comparative coefficient of variation of inter- and intra-operator variability for this ultrasound machine compared with that of manual methods has previously been reported as 8.2% and 6.4%, respectively [30].

Reference data from North American clinics

Anonymized risk factor data (see Table 1) and results from the carotid examination for 481 individuals were obtained from 2

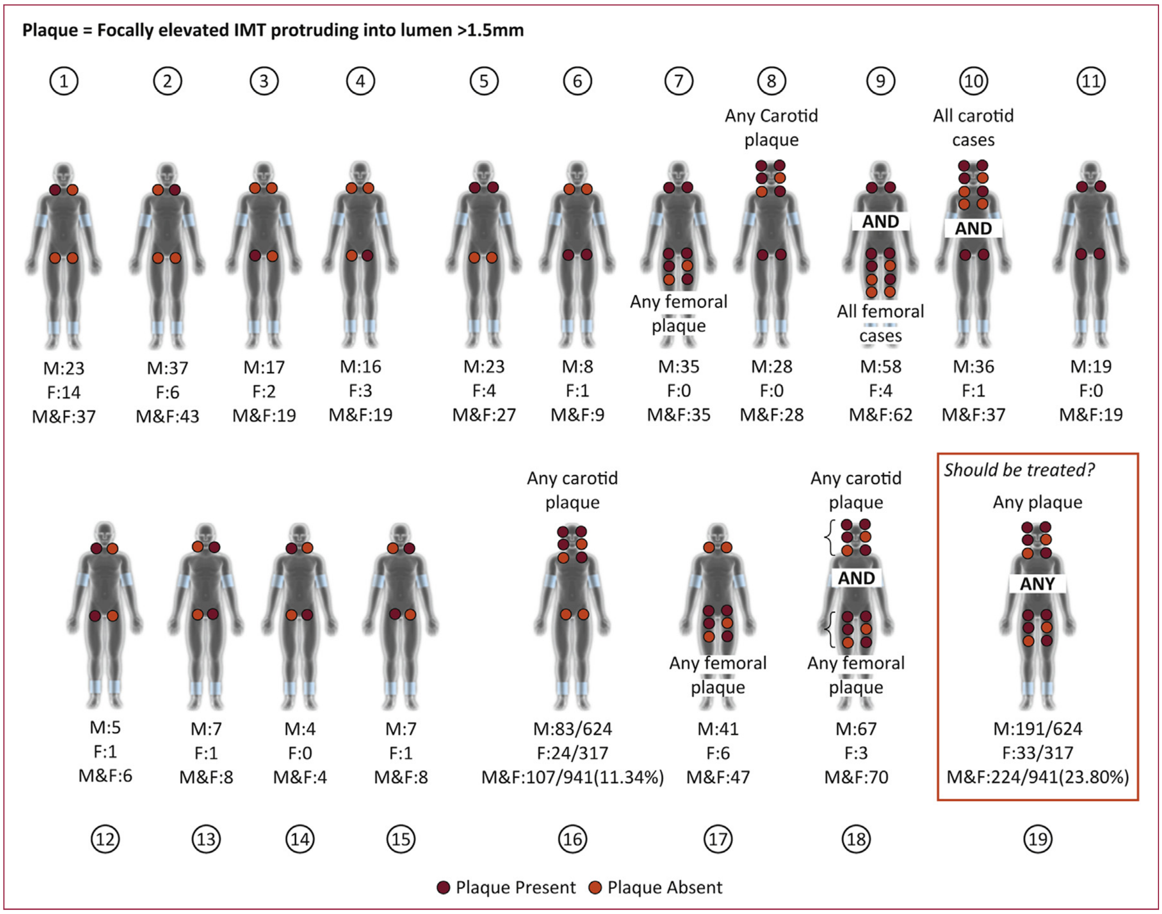

FIGURE 4. Plaque distribution by arterial site from study in India. A total of 11.3% of subjects had carotid involvement only (case ⑯); 5% had iliofemoral artery involvement only; 7.4% had both carotid and iliofemoral artery involvement; and the total number of subjects eligible for medical intervention was 23.8% (case ⑲) on the basis of ultrasound imaging. F, female; IMT, intima-media thickness; M, male.

North American clinics for comparison purposes. The average age of this cohort was 59.68 ± 11.95 years (female, 39%), which was around 15 years older than the average age of the Indian cohorts (44.27 ± 13.76 years). Mean age of the constituent cohorts from Toronto and Richmond was 53.63 ± 9.23 and 65.32 ± 11.42 years, respectively. The number of current smokers was identical to the male cohort of former smokers from Sirsa (15%). The number of individuals on medication to control hypertension was higher (44% vs. 19% in Sirsa and 21% in Jaipur). Similarly, the number of individuals on medication to control type II diabetes in the North American cohort was higher (14% vs. 2% in Sirsa, 6% in Jaipur). Overall, 42% of the cohort from North America exhibited carotid plaque presence compared with 19% of the cohorts from India. Four asymptomatic patients from Texas were referred to a specialist clinic for additional studies, which led to a carotid endarterectomy procedure in all 4.

Risk profile data were used to establish eligibility for lipid-lowering therapy based on National Cholesterol Education Program ATP III (Third Report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults, Adult Treatment Panel) guideline [16] and 2013 ACC/AHA ATP IV criteria [31,32]. A comparison of eligibility based on the 2 guidelines is illustrated in Figure 6, which shows that the number of individuals who would qualify for medical intervention under ATP III guidelines was 71. Of these, only 37 had carotid plaque, which means that 166 individuals with carotid plaque (34% of total cohort) did not qualify for treatment under ATP III. The number of individuals that qualify for medical intervention under ATP IV guidelines increased from 71 to 217. Of these, only 136 had carotid plaque; a total of 67 individuals with carotid plaque (14% of total cohort) did not qualify for treatment under ATP IV. In the lowest ATP III risk category (Framingham Risk Score <10%, risk factors <2, and low-density lipoprotein <190 mg/dl), there were 65 individuals or 14% of the cohort that exhibited carotid plaque. Similarly, there were 67 individuals with carotid plaque in the lowest and second-lowest ATP IV risk categories. These data show that even in the lowest ATP risk

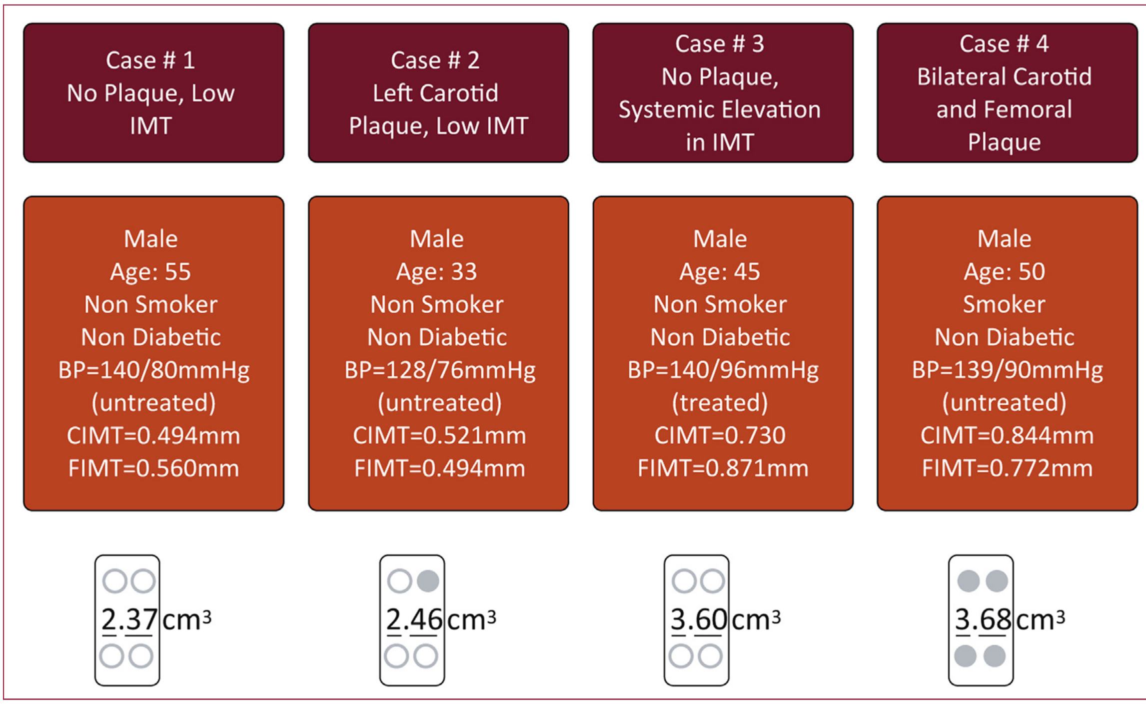

FIGURE 5. Illustrative examples of the FUN score. For illustrative purposes, preliminary FUN score on 4 asymptomatic individuals was computed. The first case involves a 55-year-old male subject with no known atherosclerotic cardiovascular disease (ASCVD) risk factors. An ultrasound examination of 4 arterial sites revealed right and left common carotid mean IMT of 0.490 mm and 0.498 mm, respectively. Mean IMT in the right and left common iliofemoral arteries was found to be 0.565 mm and 0.554 mm, respectively. Position-monitored transversal plane sweep was used to compute IMV over a 5-cm segment of each artery; the 4 IMV measurements were subsequently summed to reveal a FUN score of 2.37 cm^3 for this individual. No plaques were found in the 4 arterial beds examined for case 1 (bilateral common carotid and bilateral common iliofemoral arteries and their bifurcations). Case 2 involves a 33-year-old man with no ASCVD risk factors, but who was found to have a plaque in the bulb of the left carotid. Case 3 involves a 45-year-old man undergoing treatment to control hypertension but no other ASCVD risk factors. In this case, a systemic elevation in IMT was seen in all 4 arterial beds, but no plaques were found. Finally, case 4 involves a 55-year-old man who had no ASCVD risk factors other than smoking. In this individual, plaques were found in all 4 arterial beds examined but the FUN score was almost the same as that for case 3 (who had no plaques). This observation illustrates the importance of separately notating plaque findings as illustrated. BP, blood pressure; CIMT, carotid intima-media thickness; FIMT, femoral intima-media thickness; other abbreviations as in Figures 1 and 2.

categories, about 15% of the cohort have carotid plaque; these patients would otherwise be missed by the U.S. guidelines for treatment. Though the number of individuals with plaque that qualify for treatment under ATP IV increased from 37 to 136, the number of individuals without plaque that qualify for treatment under ATP IV also increased substantially (from 7% to 17% of the cohort). The effect of implementing the ATP IV guideline could potentially lead to overtreating a sizable subset of individuals with lipid-lowering agents who may not necessarily have progressed to developing atherosclerotic disease.

DISCUSSION

Our study shows that automation in ultrasound imaging technology allows even nonexpert users to evaluate the presence of subclinical atherosclerosis. Detection of subclinical disease is further enhanced by inclusion of the iliofemoral artery examination. We have shown that quantification of overall atherosclerotic disease burden in the peripheral arteries is feasible by measuring IMV and summing the results in a simple index (FUN score). Our results showed that ultrasound examination, when compared with ATP III and ATP IV guidelines, allowed improved identification of individuals who could be targeted for prophylactic medical intervention.

No reliable data about traditional ASCVD risk factors were available for the cohorts studied in India. Therefore, this study provided useful insights into the prevalence of subclinical atherosclerosis in a population where the norms for biochemical profiles did not exist [4]. Focusing just on the male subjects, prevalence of

TABLE 1. Baseline observations for reference data from North America

	N = 481
Age, yrs	59.68 ± 11.96
Male	294 (61)
Diabetes mellitus	65 (14)
Current smoker	73 (15)
Systolic blood pressure, mm Hg	122.52 ± 14.53
Diastolic blood pressure, mm Hg	77.72 ± 8.18
On hypertension medication	214 (44)
LDL-C, mg/dl	121.28 ± 42.42
HDL-C, mg/dl	57.77 ± 32.48
TC, mg/dl	202.23 ± 49.12
Right CCA mean IMT, mm	0.63 ± 0.15
Left CCA mean IMT, mm	0.67 ± 0.16

Values are mean ± SD or n (%).
CCA, common carotid artery; HDL-C, high-density lipoprotein cholesterol; IMT, intima-media thickness; LDL-C, low-density lipoprotein cholesterol; TC, total cholesterol.

atherosclerotic plaques in a relatively young (49 ± 12 years old) cohort of asymptomatic urban male population from Jaipur was high (41%) compared with the overall prevalence of 24%. It should be noted that the proportion of subjects on antihypertensive medication were almost identical in both the semiurban and urban communities from India, around 20%. Similarly, the percentage of males on medication to control type II diabetes was also comparable (2% semiurban vs. 6% urban). However, there was a stark difference in the number of male smokers or former smokers (15% semiurban vs. 40% urban). This difference alone may serve to explain the higher prevalence of plaque findings in the urban cohort from Jaipur.

Plaque findings in the 2 Indian cohorts did not correlate with systolic or diastolic blood pressure. This finding is intriguing because hypertension is an important risk factor for atherosclerosis. Prevalence and mortality rates from hypertension in populations from low- and middle-income countries are known to be particularly high despite minimal atherosclerosis of the coronary arteries [33]. The overall younger age of the Indian cohorts studied here combined with hitherto unknown prevalence of peripheral arterial atherosclerosis may explain this confounding result; though a more targeted study is needed to further explore this observation.

Overall, carotid plaque presence in the reference data from North America was 42% compared with 19% of the cohort from India. Younger mean age (44.27 ± 13.76 vs. 59.68 ± 11.95 years) and the aggressive lifestyle intervention in the cohort from Sirsa may explain the overall lower incidence of plaque finding in the Indian population. However, carotid plaque prevalence in the male cohort from Jaipur (37%) was similar to the prevalence in the male cohort from the reference data from North America (41%) despite the fact that the mean age in Jaipur was lower by 9 years. This may be related to earlier prevalence of disease in South Asians [34] and the resulting alarming rise in ASCVD-related complications and mortality rates in India [35]. It seems that plaque information from ultrasound images may serve as a guide for initiating medical intervention regardless of the availability or knowledge of traditional risk factors. Our results further suggest that not only in low- and middle-income countries, but even in the developed nations, ultrasound images may help refine strategies for medical intervention. It will however still be too contentious to suggest that risk factors—positive and imaging-negative asymptomatic subjects may be spared from medical intervention. Conversely, arguments against initiating medical intervention on risk factors—negative and imaging-positive asymptomatic subjects become harder to justify.

From data collected in developed nations, Sillesen et al. [21] have also recently reported carotid plaque burden as a new measure of subclinical atherosclerosis based on a bilateral examination of the extracranial carotid system with 3D ultrasound. Carotid plaque quantified by this method was shown to be better related to coronary artery calcium score than ankle brachial index, abdominal aortic diameter, and IMT of the common carotid (n = 6,101, mean age 68.8 years). It was suggested that carotid plaque burden will lead to improved prediction of ASCVD-related events and that it may detect early stages of disease even before coronary calcification. This is particularly relevant to the present study because the average age of the cohort examined from India was around 44 years. Alternative tests such as ankle brachial index and abdominal aortic diameter represent a much severer disease manifestation at an older age. Furthermore, in a relatively young cohort that includes pre-menopausal women, a coronary calcium score is neither desired nor recommended. The improvised imaging technique employed in the present study included transducer positioning data to address quantification errors from image slice misalignment and unknown interslice interval. In addition, the FUN score provides a comprehensive and standardized index of subclinical atherosclerosis by adding bilateral examination of the iliofemoral artery and by analyzing data from a standardized segment of each of the 4 arteries examined. The systemic nature of atherosclerotic disease is such that various vascular beds develop atheroma at different stages of severity, including calcium deposits [36]. Based on the 10-year observational study reported by Belcaro et al. [23], evaluation of a greater number of vascular beds incorporated in the FUN score may allow higher specificity for the detection of subclinical atherosclerotic disease. Our study has shown that modesty is not a major barrier to include iliofemoral scanning even in conservative communities. Only 2 females declined consent from the cohort examined in Jaipur.

Three-dimensional ultrasound is an emerging field and its most fundamental role will likely be to automate plaque detection so as to enable less qualified personnel to

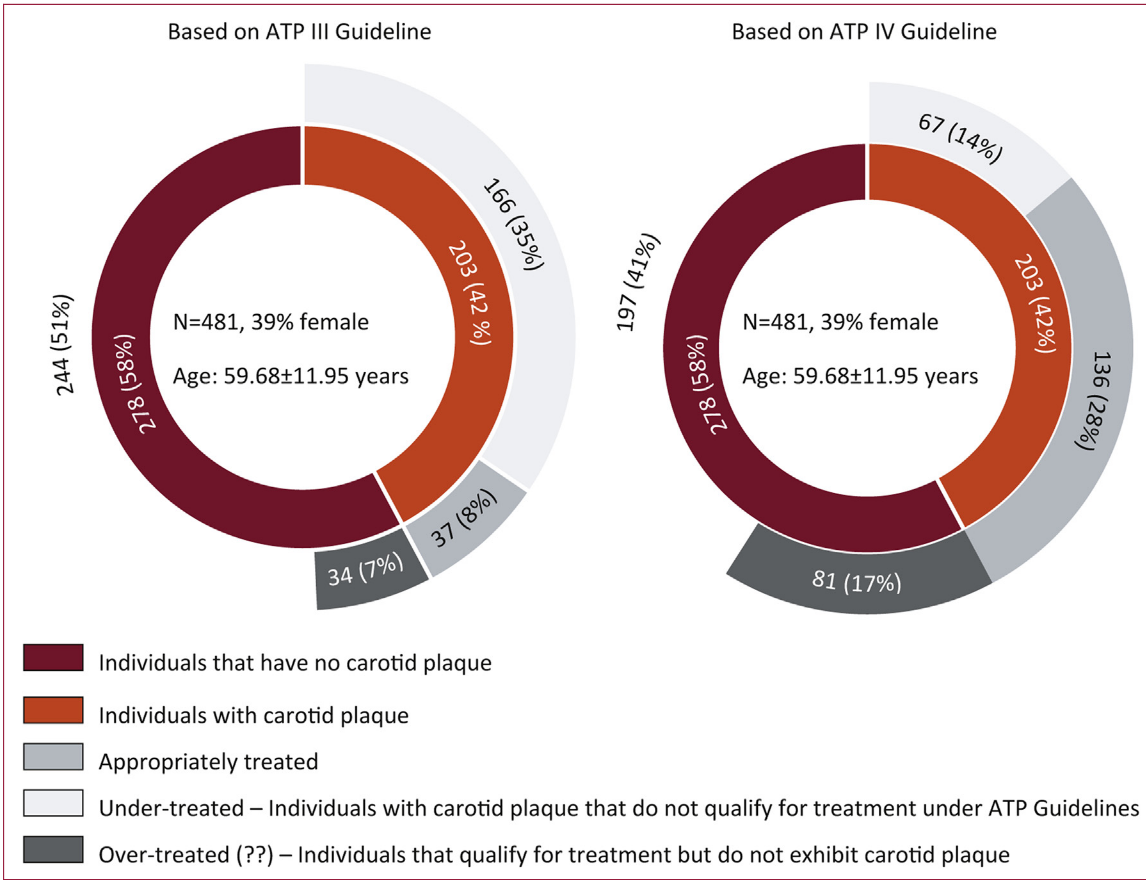

FIGURE 6. Plaque findings and eligibility for medical intervention based on ASCVD prevention guidelines. Comprehensive atherosclerotic cardiovascular disease (ASCVD) risk factor data for 481 self-referred patients analyzed from 2 clinics in North America showed that 35% of the cohort with carotid plaque would have failed to qualify for medical intervention based on the National Cholesterol Education Program ATP III (Third Report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults [Adult Treatment Panel]) guideline. Under the recently introduced American College of Cardiology/American Heart Association ATP IV prevention guideline, 14% of the cohort with carotid plaque would have been ineligible for lipid-lowering treatment.

administer the examination. The resulting plaque quantification may become clinically relevant for monitoring response to medical intervention. However, for making the initial clinical decision about whether to initiate medical intervention or not, the simple binary approach of plaque presence or absence from a 2D B-mode ultrasound examination may be adequate. Our data demonstrate that this approach may detect many subjects who may not have been picked up by routine risk factor profiling. However, uncertainty may still persist in subjects who have elevated risk factors (when available) but the imaging studies are normal.

Study limitations

The present study did not try to identify the relationship of carotid and iliofemoral artery involvement with coronary calcification or coronary events. Neither was an attempt made to readdress the relationship of pre-clinical atherosclerosis and the prevalence of standard biochemical and constitutional risk factors in the cohorts from India. The present study was designed to evaluate the feasibility of assessment of subclinical atherosclerosis at the population level in more than solitary peripheral arterial beds to identify those subjects that might benefit from prophylactic medical intervention. The latter objective is commensurate with the primary ATP IV recommendation for the eligibility of preventative medical intervention. This study was cross-sectional in nature and by itself does not have the ability to compare prognostic value of atherosclerotic plaque findings to future ASCVD events. Traditional risk factor assessment was not completed primarily because of the unavailability of serum markers during the medical camps held in India especially due to cost constraints; the imaging endeavor was entirely based on uncompensated time volunteered by a group of radiology residents and cardiologists. The simple questionnaire used for collecting information about smoking history, family history of ASCVD and medications for hypertension and diabetes may not have been fully understood by the enrollees. The ultrasound examination was administered in an exceedingly fast-paced environment (typical exam time <5 min) by radiology residents (trained on the spot) to demonstrate the feasibility of the technique. Plaque search was performed manually using B-mode ultrasound images and may have been missed by the novices [37]. Therefore, potential false-positive and false-negative findings in this study are not known. A single-angle IMT measurement was taken in the far wall of the common carotid and far wall of the common femoral artery. It is possible that a higher IMT may have been found at an alternative scanning angle. However, given the younger average age of the cohorts examined in India, it is unlikely that the error exceeds the resolution of B-mode ultrasound images. This assumption is based on an earlier unpublished investigation of 30 relatively young (average age around 40 years) volunteers whose far-wall common carotid IMT was measured at multiple angles. Study sites were selectively chosen; therefore, the results of this study cannot be generalized to a more diverse population. Finally, automated image slice alignment and contour detection for the computation of the FUN score was not fully functional at the time of image acquisition, necessitating manual adjustments. Preliminary intraobserver error in the FUN score calculation was estimated to be 30%. This is comparable to the error generally accepted for the computation of coronary artery calcium score using the Agatston method. However, improvements to the FUN score accuracy are needed along with clinical validation of plaque findings.

CONCLUSIONS

B-mode ultrasound is a safe, quick, easy to perform, noninvasive, and widely available imaging tool that may help in better identifying those individuals who would benefit from medical intervention for primary prevention of ASCVD-related events. Automation and overall image improvements over the past decade have made this technique clinically viable. Even though the updated ACC/AHA guidelines recommend against carotid IMT testing [31], the panel drafting this document did not consider plaque findings in their recommendation. Clearly, direct evidence of subclinical atherosclerotic plaque burden incorporated in a simplified and standardized FUN score index provides motivation for aggressive medical intervention. In developing countries such as India where a much younger population is afflicted with the devastating consequences of ASCVD-related events, an alternative strategy for identifying high-risk individuals may not be out of place.

This study shows that rapid screening for subclinical atherosclerosis is feasible with automated ultrasound examination of carotid and iliofemoral arteries. Adding B-mode examination of the iliofemoral arterial beds identifies additional individuals who would benefit from prophylactic medical intervention for the prevention of ASCVD-related events. Surely, such a simple approach merits adoption on a wide scale as a modern approach to a modern scourge of rapidly rising ASCVD-related events worldwide. Furthermore, the lower incidence of plaque presence in a more disciplined cohort reported here should inspire the promotion of cardiovascular health by lifestyle modification.

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