The Chennai Urban Rural Epidemiology Study (CURES) - Study Design And Methodology (Urban Component) (CURES - I)

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Abstract

The report of World Health Organization (WHO) shows that India tops the world with the largest number of diabetic subjects. This increase is attributed to the rapid epidemiological transition accompanied by urbanization, which is occurring in India. There is very little data regarding the influence of affluence on the prevalence of diabetes and its complications particularly retinopathy in the Indian population. Furthermore, there are very few studies comparing the urban / rural prevalence of diabetes and its complications. The Chennai Urban Rural Epidemiology Study (CURES) is designed to answer the above questions. CURES is initially planned as a cross-sectional study to evolve later into a longitudinal study. Subjects for the urban component of the CURES have been recruited from within the corporation limits of Chennai City. Chennai (formerly Madras), the largest city in Southern India and the fourth largest in India has been divided into 10 zones and 155 wards. 46 wards were selected by a systematic random sampling method to represent the whole of Chennai. Twenty thousand and one individuals were recruited for the study, this number being derived based on a sample size calculation.

The study has three phases. Phase one is a door to door survey which includes a questionnaire, anthropometric, fasting capillary blood glucose and blood pressure measurements. Phase two focussed on the prevalence of diabetic complications particularly retinopathy using standardized techniques like retinal photography etc. Diabetic subjects identified in phase one and age and sex matched non-diabetic subjects will participate in these studies. Phase three will include more detailed studies like clinical, biochemical and vascular studies on a sub-sample of the study subjects selected on a stratified basis from phase one. CURES is perhaps one of the largest systematic population based studies to be done in India in the field of diabetes and its complications like retinopathy, nephropathy and neuropathy.

INTRODUCTION

Epidemiology is defined as “the study of the distribution and determinants of health related status or events in specified populations”.1 Epidemiological studies highlight the health issues currently affecting the nation and also help us to delineate the risk factors associated with the disease. Furthermore these studies also guide us to estimate the disease burden of the future, which is essential to plan preventive health strategies.

Currently, India is passing through an epidemiological transition due to rapid urbanization coupled with economic growth.2 The changing pattern in the economy is obvious from the current urbanization rate which stands at 35% compared to 15% in the 1950’s. This rapid transition will have a major implication on the present and future disease patterns in India with particular reference to non-communicable diseases like diabetes and coronary artery disease (CAD).3,4

Both CAD and diabetes are consequences of the insulin resistance syndrome (IRS) also known as the metabolic syndrome. This is a cluster of metabolic abnormalities comprising of abdominal obesity, glucose intolerance / type 2 diabetes mellitus, dyslipidemia and hypertension.5 Our earlier population based study the Chennai Urban Population Study (CUPS) showed intra-urban difference in the prevalence of various components of the IRS.6 This study also reported on the prevalence of hypertension, dyslipidemia, CAD, peripheral vascular disease (PVD) and diabetic retinopathy.6-8 However, the limitation of the CUPS was that it was conducted in a selected population in Chennai representing the middle and low-income group. This restricts extrapolating the obtained results to whole of population of Chennai. Moreover the study numbers being small (n=1262), CUPS was underpowered.

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to answer several questions particularly regarding prevalence of complications of diabetes like retinopathy, neuropathy and nephropathy as there were only a total of 152 diabetic subjects in CUPS. Finally there was no rural component.

Furthermore, there is paucity of data on the prevalence of diabetic retinopathy in native Indians. The available studies indicate the prevalence to be much lower than that compared to Europeans.9 The reason for this decreased prevalence of diabetic retinopathy in contrast to the increased diabetes prevalence in Indians is not very clear.

This forms the background for undertaking a study called the “Chennai Urban Rural Epidemiology Study” (CURES), which is planned to be a large cross-sectional study, of representative samples of the whole of Chennai (representing the urban component) and villages around Chennai (representing the rural component).

The CURES study commenced in August 2001 with the objective of comparing the prevalence of the various components of IRS in an urban and rural South Indian population and also to assess the prevalence of diabetes-associated complications, particularly retinopathy and disorders like glaucoma and cataract in Type 2 diabetic subjects. This paper will discuss the study design and methodologies adopted for CURES in the urban component of the study.

Study design

CURES is planned as a cross-sectional field survey, which measures the prevalence of disease. This design has been adopted by several countries on representative samples of their populations focusing on personal and demographic characteristics, illnesses and health related habits. According to the World Health Organization (WHO), in epidemiological research, cross-sectional field survey is the first step to obtain accurate baseline values to later plan a prospective follow up study.

Sample size calculation

Earlier published studies on selected populations in India have suggested that the prevalence of known diabetes in urban areas is around 5.0%.6,10 If we are to assess the risk factors of diabetic retinopathy, we require a minimum of 200 subjects affected by retinopathy. Based on the prevalence rate for diabetic retinopathy obtained in CUPS in known diabetes (21.4%),8 we require 1000 diabetic subjects in order to have 200 subjects affected by diabetic retinopathy. To obtain 1000 adult (≥20 years) diabetic individuals, a sample size range between 16,000 to 24,000 was calculated assessing 99% confidence intervals and 0.5% error. The upper limit of 24,000 was taken as our target. However, considering a drop out rate of 10% among the diabetic subjects, we decided to recruit 26,000 individuals for the study.

Sampling design

Chennai (formerly Madras), the largest city in Southern India and the fourth largest in India is located on the Coromandel coast of the bay of Bengal and has a population of 4,216 million.11 The whole of Chennai has been divided into 10 zones and 155 wards by the Chennai Corporation. The sampling for CURES was based on the model of systematic random sampling, wherein, of the 155 wards, 46 wards were selected to represent all the 10 zones. The total sample size of 26,000 individuals was selected from these 46 wards. The sample distribution in each ward within these zones is based on the proportion of their population in that particular zone. The wards selected for the study are shown in Fig. 1. Further, within each ward, every third lane or road, following the right hand rule was surveyed. Such a sampling approach was chosen as it enabled the arrival of an equitable distribution of the entire Chennai population while ensuring that the sampling error is kept to a minimum. Another advantage is the simplicity of the administrative procedures involved. All men and women ≥ 20 years of age were considered eligible for the study.

**Phases Of The Study:**

CURES was conducted in three phases (Fig. 2):

Phase one was the main survey conducted in the field, which involved a door-to-door survey in the selected wards.
Phase two, a detailed study of the vascular complications, particularly retinal photography and detailed examination of the eye in all diabetic subjects identified in Phase 1 with age and sex matched non-diabetic subjects as controls.

Phase three aimed at studying the normal distribution of various measures inclusive of biochemical parameters, obesity and anthropometric indices, atherosclerotic markers like carotid intimal medial thickness and detailed eye examination to assess eye disorders in a sub-sample of the total population recruited for CURES.

**Phase I Of The Study**

The first phase of the study was done with the help of an external agency. This phase included training of survey team, obtaining the consent of the study individuals and finally conducting of survey itself (Fig. 3).

**a. Intensive training programme:**

Training was provided on a one to one basis to all the 50 team members and lasted for 5 weeks consisting of training sessions of 2-3 hours per day on all working days of the week. The training programme was conducted by the epidemiology team of the Madras Diabetes Research Foundation before the survey commenced. The aim was to ensure that each member of the team was well trained in various procedures which included filling out the questionnaire which has been validated and various anthropometric, fasting blood glucose and blood pressure measurements. Anthropometric measurements included height, weight, waist, hip and mid-arm circumference measurements. The trainees were educated to use the electronic machine to measure blood pressure and the glucometer for estimating fasting capillary blood glucose. The main objective of this training was to avoid biases or errors in any of the procedures employed.

Each trainee was evaluated individually. SS and DM took turns and cross-verified and validated the interview and measurements done by the trainees. The trainees were considered fit to conduct the study only if they achieved minimum error rates for all the measurements.

**b. Quality controls:**

The machines used for the study namely the electronic BP apparatus and glucometers were validated. The glucometer was calibrated every day with a calibrator provided along with the machine. Reproducibility for the glucometer was assessed by measuring blood glucose for the same patient six times as well as by evaluating the machines against each other. The machines were used for the study only if the coefficient of variation were <5%. Similar procedure was followed for the blood pressure machines.

Selected questions from the questionnaire were used for validation purposes. These included questions related to knowledge on diabetes, medical history and family history of diabetes. The questionnaire was retested on 50 subjects after 5 weeks and correlation analysis revealed good reproducibility. Monitoring the persons conducting the survey was done randomly rechecking the data and values, with particular reference to blood pressure and fasting blood sugar estimations.

**c. Pilot study:**

Initially, a pilot study was conducted among a sub-population of 100 subjects to overcome the practical difficulties of performing the study. The study was conducted as a four-step procedure. The first step was selection of the participants. Next the informed consent was obtained after which an interview was conducted and finally fasting capillary
blood glucose and blood pressure was measured. Five teams with four members each conducted the pilot study. The teams visited selected households one day in advance to request eligible individuals to observe a minimum of 8 hours fast prior to estimation of the fasting capillary blood glucose levels the next morning. The pilot study revealed that it took 45 minutes to collect all the information, which included the interview (25 minutes), anthropometric measurements (5 minutes) and measuring fasting capillary blood glucose levels and blood pressure (15 minutes). The pilot study concluded that there was no questionnaire fatigue and a good response rate provided the confidence to proceed with the study.

d. **Obtaining consent and ethical committee approval:**

Ethical committee approval was obtained prior to the start of the study and an informed consent was obtained from all the study subjects. Each step in the study was verbally explained to the participant and adequate opportunities were given for discussion of questions with the interviewer. A written description of the study was shown to the participant for obtaining his/her consent. The consent form was translated into the regional language (Tamil) for the convenience of individuals who did not know English. It was also back translated to check the accuracy of the translation. The consent form was explained to each participant and consent was obtained only after the interviewer was sure that the participant understood and accepted the contents.

e. **Method of the Survey:**

The main study was conducted using the four step procedure as adopted in the pilot study.

Questionnaire: The questionnaire which was administered to the study subjects included details regarding demographic and socioeconomic characteristics like participant’s self-reported age, sex, educational status, family and individual income, type of house, etc. Behavioural aspects included self-reported current and past smoking and alcohol use. Health awareness included knowledge regarding prevalence, risk factors and prevention of diabetes. Health conditions were documented based on self-reported history of diabetes, hypertension and cardiovascular disease (chest pain, heart attack and stroke). Family history for all of the above conditions was also collected. Health economics data included the amount spent on health and details of hospitalization for the past five years. Details pertaining to psychosocial aspects like depression, stress and hopelessness were obtained using standard instruments. Physical activity was assessed by using a standard scale, which was validated earlier in a South Indian population.12

**Anthropometric measurements**

Height was measured with a tape to the nearest cm. Subjects were requested to stand upright without shoes with their back against the wall, heels together and eyes directed forward.

Weight was measured with traditional spring balance that was kept on a firm horizontal surface. The scale was checked every day and calibration was done with “known” weights. Subjects were asked to wear light clothing and weight was recorded to the nearest 0.5 kg.

Body mass index (BMI) was calculated using the formula: weight (Kg)/height (m²).

Waist was measured using a non-stretchable fibre measure tape. The participants were asked to stand erect in a relaxed position with both feet together, one layer of clothing was accepted. Waist girth was measured as the smallest horizontal girth between the costal margins and the iliac crests at minimal respiration.

Hip was taken as the greatest circumference at the level of greater trochanters (the widest portion of the hip) on both sides. Measurements were made to the nearest centimeter.

Waist and hip ratio (WHR) was calculated by dividing the waist circumference (cm) by the hip circumference (cm).

Blood pressure and blood glucose measurement: Blood pressure was recorded in the sitting position in the right arm to the nearest 1mm Hg using the electronic OMRON machine (Omran Corporation, Tokyo, Japan). Two readings were taken 5 minutes apart and the mean of the two was taken as the blood pressure. Fasting capillary blood glucose was determined using the glucometer (Lifescan Johnson & Johnson, Milpitas, California, USA).

**Definitions of variables used in Phase I of CURES:**

- Diabetes was diagnosed based on the past medical history, drug treatment for diabetes, and/or using the ADA fasting criteria.13
- Current age was defined as the age at the time of examination (2001 - 2002).
- Duration of diabetes was taken as the difference between the date the individuals were first told by a medical practitioner that they had diabetes or the first time their fasting blood sugar level met the ADA diabetes diagnostic criteria or when the hyperglycaemic medications were started to the date of the examination.
- Hypertension was diagnosed based on past medical history, drug treatment for diabetes, and/or if the subjects had systolic blood pressure (SBP) of 140 mmHg or greater and/or diastolic blood pressure (DBP) of 90 mmHg.

**PHASE II OF CURES**

From the Phase I of the study, all the known diabetic subjects and age and sex matched non-diabetic subjects were invited to the centre for detailed studies on various vascular complications (the Diabetes Complication Study). In Phase 2 the study subjects were requested to undergo detailed anthropometric measurements, complete biochemical tests and the special tests (Fig. 4). Informed consent was once again obtained from every participant to undergo this phase of the study.

**Anthropometric measurements**

Weight, waist and hip were again measured using methods as mentioned in Phase 1.
Skin fold measurement

All the measurements were taken in duplicate on the right side of the body with the subject lightly clothed. Measurements were taken to the nearest millimeter using Lange skinfold calipers (Cambridge Scientific Industries, Inc, Cambridge, Maryland). The sites of measurement were biceps, triceps, subscapular, chest, supra-iliac, mid-axillary, abdomen, mid-thigh and medial calf using standard techniques.15

The skin was firmly grasped by the thumb and index finger of the left hand about 1cm or 1/2 inch proximal to the skinfold site and was pulled away from the body. The caliper held in the right hand was placed perpendicular to the long axis of the skin with the caliper’s dial facing up so that it was easily readable. The caliper tips were placed on the site of measurement about 1cm or 1/2 inch distal to the fingers holding the skin, so that pressure from the fingers did not affect the measured value.

Triceps: The triceps skinfold thickness was measured on the posterior aspect of the right arm, over the triceps muscle, midway between the lateral projection of the acromion process of the scapula and the inferior margin of the olecranon process of the ulna.

Biceps: The biceps skinfold is a vertical fold on the anterior aspect of the arm, over the belly of the biceps muscle, directly opposite to the triceps skinfold site.

Chest: The chest or pectoral skinfold measurements were made using a skinfold with its long axis running from the top of the anterior axillary fold to the nipple. The skinfold was grasped as high as possible at the anterior axillary fold, and the thickness of the fat fold was measured 1cm or 1/2 inch below the fingers along the axis.

Subscapular: The subscapular thickness was measured 1cm below the inferior angle of the scapula, which was identified by gentle palpation of the lower end of the scapula with the patient standing with his arms by his side.

Midaxillary: This was measured at the level of the right midaxillary line: (where it is met by a horizontal line from the xiphisternum (at the bottom of the sternum where the xiphoid process begins). It was measured with the subject standing erect with the right arm slightly abducted and flexed (bent posteriorly).

Suprailiac: This skinfold was measured just above the iliac crest in the midaxillary line.

Abdomen: The study subjects were asked to stand erect with their body weight evenly distributed on both feet. They were then asked to breathe evenly and relax their abdominal muscles. A horizontal fold of skin 1 cm below the umbilicus and 3 cm laterally (to the right) was then measured.

Thigh: This was measured as vertical skin fold in the anterior aspect of the thigh midway between the inguinal crease and the upper border of the patella in the midline. Flexing the subject’s hip helped to locate the inguinal crease.

Medial calf: The point of maximum calf circumference was marked at the medial (inner) aspect of the calf. A vertical skin fold was grasped about 1 cm proximal to the marked site and measured at that site.

Sagittal abdominal diameter: Abdominal height (sagittal abdominal diameter), defined as the width of the abdomen at the waist level, was measured with a sliding beam abdominal caliper (Holtain-Kahn abdominal caliper; Holtain, Dyfed, Wales) with the subject lying supine on the examination table.16

Biochemical tests

Fasting and 2 hour post-load (post 75-g oral glucose load - oral glucose tolerance test [OGTT]) plasma glucose concentrations were done in all subjects except in known diabetic subjects. Biochemical analysis was done on a Hitachi - 912 Autoanalyser (Hitachi, Germany) using kits supplied by Roche Diagnostics (Basel, Switzerland). Fasting plasma glucose (GOD - POD method), serum cholesterol (CHOD-PAP method), serum triglycerides (GPO-PAP method) and HDL cholesterol (direct method) were also measured. Glycated haemoglobin (HbA1C) was estimated by high-pressure liquid chromatography using the Variant machine (Bio-Rad, Hercules, Calif., USA). Serum insulin concentration was estimated using Dako kits (Dako, Glostrup, Denmark).

Special investigations

Doppler:

The diagnosis of peripheral vascular disease was based on palpation of foot pulses, clinical symptoms of claudication and measurement of an ankle-branchial index (ABI). This included recording of pressure tracings using the KODY.
Vaslab Machine (Kody Labs, Chennai, India). Blood pressure recordings were made of the brachial pulses in the upper limb. Similar recordings are made of the dorsalis pedis and posterior tibial pulses in the lower limb by inflating the cuff proximal to the ankle and the mean of these two readings was taken as the ankle pressure. An ABI index of less than 0.9 was the criteria used for the diagnosis of peripheral vascular disease.17

**Biothesiometry:**

A biothesiometer (Biomedical Instrument Co., Newbury, Ohio, USA) was used to assess vibratory perception threshold (VPT) of the great toe in a standardized fashion. Subjects were initially requested to remove their shoes and socks and lie supine on a couch for at least 5 minutes before the measurements were done. The foot was kept warm during the measurement and as the room was air-conditioned, the temperature of the room was around 25°C. The biothesiometer tactor, which vibrates at 100 Hz with an amplitude proportional to the square of the applied voltage was applied perpendicular to the test site with a constant and firm pressure. Subjects were initially familiarised with the sensation by holding the tactor against the distal palmar surface. VPT was then measured at the distal plantar surface of the right great toe. The voltage was slowly increased at the rate of 1 V/s and the VPT was defined as the moment when the subject indicated he / she first felt the vibration. The voltage at which this occurred was recorded. Three further cycles were performed and the average value was taken.

Neuropathy was diagnosed using a neuropathy scoring system (NSS), which included symptom evaluation, tendon reflexes and sensory tests in addition to the biothesiometry readings.

**Carotid Doppler:**

The intimal as well as medial thickness of the carotid arteries were determined using a high resolution B mode ultrasonography system (Logic 400 GE, Milwaukee, Wis., USA) having an electric linear transducer midfrequency of 7.5 MHz. The images obtained were recorded and photographed. The scanning was done for an average of 20 min. The intimal plus medial thickness (IMT) was measured as the distance from the leading edge of the first echogenic line to the second echogenic line. The first echogenic line represented the lumen intimal interface and the second line was produced by the collagen - containing upper layer of the intimal adventitia. Six well defined arterial wall segments were measured in each (right and left) carotid system: the near wall and far wall of the proximal 1 cm of the internal carotid artery, the near wall and far wall of the carotid bifurcation, beginning at the tip of the flow divider, and extending 1 cm below this point, and the near wall and far wall of the arterial segment, extending 1 cm below the tip of the flow divider into the common carotid artery. The reproducibility of the IMT measurement was examined by conducting another scanning 1 week later on 20 subjects by same sonographer. Correlation analysis revealed a fairly good correlation between the IMT measurements.

**Electrocardiogram:**

A resting 12-lead electrocardiogram (ECG) was carried out on all subjects. Minnesota coding was used to grade the ECG. ECG changes suggestive of ST-segment depression (Minnesota codes 1-1-1 to 1-1-7) or Q-wave changes (Minnesota codes 4-1 to 4-2) or T-wave changes (Minnesota codes 5-1 to 5-3) were taken as evidence of coronary artery disease.

**Studies on ocular changes in diabetes**

A comprehensive ocular examination was done in all study subjects by trained optometrists and technicians.

Visual acuity was recorded by trained optometrists using an illuminated Snellen’s chart. The presenting and best-corrected visual acuity was documented separately for each eye.

A complete slit lamp examination was done and the depth of anterior chamber was assessed using Van Herrick’s technique, Cataract if present was documented as cortical lens changes greater than 3 and / or post-subcapsular cataract > 2 and / or nuclear opalescence > 3 (Lens Opacity Classification System) - [LOCIS].18

The intra-ocular pressure (IOP) was measured using Goldman applanation tonometer. The average of three independent readings was taken as the final intra-ocular pressure of each eye.

Gonioscopy was done using the Sussman’s hand held four-mirror gonio lens to assess the angle structures after instilling one drop of 4% xylocaine to each eye. The angle grading was done in each eye from Grade 0 - 4 using Schafer’s method for grading of the angles.

Automated full threshold visual fields were done using the Humphrey visual field analyzer to check for any visual field loss by using 24 - 2 SITA standard field chart with appropriate near correction. If the visual field obtained was unreliable or abnormal, it was repeated subsequently when the subject was relaxed.

**Corneal aesthesiometry:**

The corneal thickness was measured using a Cochet Bonnet aesthesiometer. The aesthesiometer filament was fully extended to 60 mm. The tip of the fiber was steadily advanced towards the cornea. When the end-plate of nylon filament was found to be in contact with cornea a mild pressure was exerted such that fiber had the slightest bend just visible. The response was assessed either by subjective response of patient or by objective blinking or withdrawal response. If there was no response, the fiber length was shortened in steps of 5 mm each time and procedure was repeated till a response was elicited. At times ‘blanks’ were given to test patients reliability and only reliable data was included.

**Retinal photography:**

Fundus photography was done to evaluate changes in the retina and the optic disc. The pupils were dilated using one drop each of phenylepherine 10% and tropicamide 1% into both eyes and the drops were repeated until the best
possible mydriasis was obtained. Four field colour retinal photography was carried out by a trained photographer with a Zeiss FF 450 plus camera using 35 mm colour transparencies. The four fields taken were stereoscopic pictures of the macula, disc superior temporal, and inferior temporal quadrants. If photography of any particular eye or any field was not possible due to inadequate dilatation, inability to cooperate properly or opacities of the media, then these were specified as missing eye or fields.

Grading of retinal photographs: The photographs were coded using an identification number and assessed in a masked manner in order to minimize any possible bias. The photographs were graded against standard photographs of the Early Treatment Diabetic Retinopathy Study (ETDRS) grading system for severity of retinopathy.19

**Definition of retinopathy:**

Levels of retinopathy based on presence and severity of lesions.

- Level 10: No retinopathy
- Level 20: Presence of definite microaneurysms only.
- Level 30 - 51: Lesions of non-proliferative retinopathy ordered by severity.
- Level 60 - 85: Proliferative retinopathy also ordered by severity

In addition to grading to severity of retinopathy presence of clinically significant edema was also determined.19 Photographs were assessed and the study subjects were assigned retinopathy level. The final diagnosis was determined by the grade of the worse eye.

**Optic disc evaluation:**

Stereo photographs of the disc was assessed for glaucomatous damage in the study subjects.20 The photographs of the optic disc were mounted against an illuminated background and assessed with Dodson’s viewer.

A diagnosis of primary open angle glaucoma (POAG) was made if the following criteria were present:

- Vertical cup : disc ratio of 0.5 or more which corresponds to the visual field defect
- Cup disc : asymmetry of 0.2 or more between the two eyes
- Thinning of the neuroretinal rim
- IOP of more than 21 mm Hg

Normal tension glaucoma was diagnosed when the above criteria for POAG were present with a normal IOP (21 mm Hg or less).

Primary angle closure glaucoma was diagnosed if narrow or occludable angles were present on gonioscopy with visual field defects and optic disc changes.

Ocular hypertension was defined as IOP > 21 mm Hg in either eye on applanation tonometry without any visual field or disc changes of glaucoma.

**Phase III of CURES**

Every sixth subject recruited in phase I was brought to the Madras Diabetes Research Foundation. All these individuals were subjected to the clinical, biochemical, special vascular tests and detailed eye examination (Fig. 4).

**Sub-studies in CURES:**

Being a large population based study we are planning to do several sub-studies in the CURES for answering various questions related to diabetes and related disorders. The recruitment procedure for these studies may vary.

**Rural component:**

The rural component of the CURES commenced in July 2003. Rural areas have been identified using the criteria that they should be a minimum of 60 km away from Chennai. Sampling techniques will be used to recruit study subjects from these areas.

**Summary**

CURES is one of the largest systematic population-based studies on diabetes and the insulin resistance syndrome in India. Together with the rural component we expect to have a database of over 50,000 subjects representative of an urban and rural area of Chennai. Once completed, this study is expected to throw light on the prevalence and risk factors of IRS in a native Indian population. Additional insights into the effects of affluence on diabetes and associated metabolic and cardiovascular disorders will also be obtained. Finally, the study will provide data on the prevalence of retinopathy, nephropathy and neuropathy in diabetic subjects.

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REFERENCES


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