Methodology

METHODOLOGY

III

The methodology pertaining to this study entitled **"Effect of Selected Spices on Hyperlipidemic and Diabetic Adults",** carried out in two phases is presented under the following headings:

PHASE I

A. Selection of Locale and Adults

B. Eliciting the Background Information of the Hyperlipidemic and Diabetic Adults

C. Assessment of Nutritional Profile of the Selected Adults

D. Selection and Nutrient Analysis of Spices

E. Preparation of Capsules of Spice Powders and Food Costing

PHASE II

F. Conduct of the Supplementation Study

G. Evaluation of the Supplementation Study

H. Analysis of the Data

PHASE I

A. Selection of Locale and Adults

The study was carried out in and around Coimbatore city in Tamilnadu state, as the investigator is familiar with these areas and hence could establish good rapport with the people in the selected areas.

For the purpose of the survey, Housing unit and Police quarters in Gandhipuram area, Housing unit in Sivananda Colony and Kavundamplayam, residential areas of Saibaba Colony, Town Hall, R.S. Puram and Vadavalli were selected. One industrial concern namely Lakshmi Machine Works Limited,

Periyanaickenpalayam where nearly 2000 adults are employed and one Government concern Bharat Sanchar Nigam Limited (BSNL), Bharathi Park Road, where nearly 1000 adults are working were also selected. From these areas more than 1000 adults were interviewed. From this, 500 adults in the age group of 40 to 60 years with hyperlipidemia and diabetes were identified for the survey. International Diabetes Foundation estimates that most people with diabetes in low and middle income countries are middle aged (45 to 65), not elderly (65+). The controlled nature of the disease condition and their willingness to participate and co-operate in the three months feeding trial formed the basis for selection of adults. One group of 75 mild to moderate hyperlipidemic adult men was selected for the supplementation study.

In the same way, another group of 75 men who reported hyperlipidemia with symptoms of diabetes mellitus like polyuria, polydypsia, polyphagia and nocturia as identified by a physician were selected for the supplementation study after assuring their willingness and co-operation. A group of 15 adults in each category of disease was also selected as control group from the larger sample and from the same industrial concerns. Sapre *et al.*, (2000) reported that estrogen, growth hormone, corticosterols and progesterone, in females (menopausal age) have diabetogenic property which is manifold, because it increases plasma protein binding capacity of insulin thereby rendering it inactive. Hence in the present study, females were not selected and only males were included for supplementation.

B. Eliciting the Background Information of the Hyperlipidemic and Diabetic Adults

Details regarding the socio-economic characteristics including age, sex, occupation, educational status, family type, monthly income and family background were gathered through interview method using a pre-tested questionnaire (Appendix I). Interview is a research tool which involves the collection of first hand information as the interviewer is in face to face contact with the respondent. It has the added advantage of being administered to any

population and it is quite flexible (Singh *et al.*, 1994). Also, the interviewer can clear the doubts then and there and obtain the correct information.

Questions were also included to get information on their lifestyle pattern like yoga, exercise, alcohol consumption, chewing habits, smoking, dietary pattern, food intake pattern, foods included and avoided, health status like general health, history of the present condition, diabetic / hyperlipidemic trait in the family, duration and treatment of the condition, physiological symptoms experienced and other diseases if any. Trend in the consumption of spices and awareness about the spices used for the present supplementation also formed part of the questionnaire. Five hundred adults including the selected adults for supplementation study and control groups were interviewed. Interview with the selected adults was carried out at the premises of the selected industrial concerns.

C. Assessment of Nutritional Profile of the Selected Adults

For effective assessment of nutritional status a three way approach comprising of anthropometric, biochemical and dietary components should be adopted, since nutritional status cannot be determined from a single measurement alone (Sachdev and Chaudhari, 1994). In the present study nutritional assessment was carried out using anthropometric measurements, clinical examination, biochemical assessment and food and nutrient intake.

1. Anthropometric Measurements

Tests of anthropometry include measurement of body size, structure and body composition. Anthropometric measurement helps in the assessment of morphological variation, occurring due to significant functional and functional changes (Vijayaraghavan and Rao, 2001). Anthropometric measurements such as height and weight and Body Mass Index were assessed before and after the supplementation.

a. Height

In one of the concerns, namely, Lakshmi Machine Works Limited, Periyanaickenpalayam, height was recorded using the stadiometer available at the industry itself. For recording the height at BSNL, a non-flexible tape was fixed on the wall and used. All the selected adults were made to stand on a flat floor with feet parallel and back of the head and heel touching the wall. The head was held straight with arms hanging on the sides. A wooden scale was gently lowered crushing the hair and making contact with the top of the head and the height was measured to 0.1 cm accuracy. The procedure followed was based on the International Standards for Anthropometric Assessment (2001) (Plate 1).

b. Weight

Weight, in the context of human body weight measurements according to medical sciences and in sports is a measurement of mass and is thus expressed in units of mass, such as kilograms (kg). The body weight of the selected adults was taken according to the procedure given by the International Standards for Anthropometric Assessment (2001) using a portable weighing balance with minimal clothing, without foot wear, standing erect and not immediately after the meals and recorded upto an accuracy of 0.1 kg (Plate 2).

c. Body Mass Index (BMI)

The Body Mass Index (BMI) is a statistical measure of the weight of a person scaled according to height. BMI is calculated as the individual's body weight divided by the square of their height. The Body Mass Index (BMI), or Quetelet index, is a statistical measure and is frequently used to assess how much an individual's body weight departs from what is normal or desirable for a person of his or her height. The excess weight or deficiency may, in part, be accounted for by body fat although other factors such as muscularity also affect BMI. The formula universally used in medicine is a unit of measure of kg / m^2

(WHO, 2005). Body Mass Index may be accurately calculated using the formula given below:

$$BMI = Weight\ (Kg) / Height\ (m)^2$$

According to the BMI classification of World Health Organisation (WHO) (1999) the selected adults were graded as normal (18.5 to 25 kg / m^2), underweight (< 18.5 kg / m^2), over weight (25 to 30 kg / m^2) and obese (> 30 kg / m^2). BMI is meant to be used as a simple means of classifying sedentary (physically inactive) individuals with an average body composition.

d. Waist Hip Ratio (WHR)

Waist-hip ratio or Waist-to-hip ratio (WHR) is the ratio of the circumference of the waist to that of the hips. It is calculated by measuring the hip circumference at its widest part and dividing that by the waist circumference (located just above the upper hip bone).

Waist Circumference

The purpose of determining waist circumference is to gain a measure of the amount of abdominal fat (visceral fat), which has been linked to increased risk of coronary heart disease and diabetes. The waist measurement was taken as suggested by ACSM (2005) at the narrowest waist level, at the mid point between the lowest rib and the top of the hip bone (illiac crest) and the following measurements were used as reference values (Plate 3).

Risk	Men	
	Cms	Inches
Very high	> 120	> 47
High	100 - 120	39.5 – 47
Low	80 – 99	31.5 – 39
Very low	< 80	< 31.5

Hip Circumference

Hip circumference is a measure of the circumference of the hip area, i.e. the measure of the underlying hip structure, musculature and adipose tissue. When combined with the measure of abdominal girth in the Waist-Hip Ratio (WHR), it has been shown to be related to the risk of coronary heart disease. The hip measurement should be taken over minimal clothing, at the level of the greatest protrusion of the gluteal (buttock) muscles. The adult was made to stand erect with their weight evenly distributed on both feet and legs slightly parted, making sure not to tense the gluteal muscles (Welborn *et al.,* 2003). The adult was made to stand on a rigid box to make the measurement easier.

Following the above procedure and guidelines the waist and hip measurements of the selected adults were carried out and the WHR was found out (Plate 4). While recording both waist and hip measurements, it was made sure that the tape was neither too tight nor too loose, but just lying flat on the skin and was horizontal.

A WHR of 0.95 for men and 0.80 for women was shown to correlate strongly with general health (International Obesity Task Force (IOTF), 2004). WHR is often used to determine the coronary artery disease risk factor associated with obesity. If obesity is redefined using WHR instead of BMI, the proportion of people categorized as at risk of heart attack worldwide will increase threefold (Haslam and James, 2005).

2. Clinical Examination

a. Blood Pressure

The term blood pressure generally refers to arterial pressure, i.e., the pressure in the larger arteries, being the blood vessels which take blood away from the heart. Arterial pressure is most commonly measured via a sphygmomanometer, which uses the height of a column of mercury to reflect the circulating pressure (The Medical Encyclopedia, 1975).

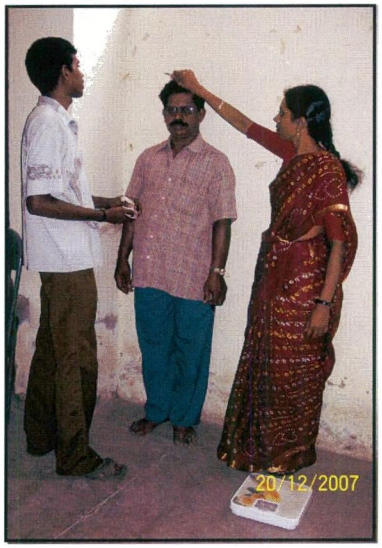

Plate 1
Measurement of Height

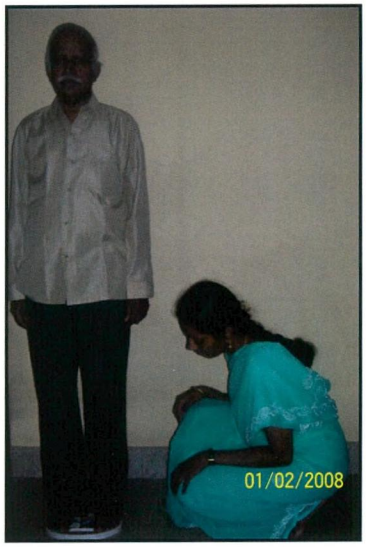

Plate 2
Measurement of Weight

Plate 3
Measurement of Waist Circumference

Plate 4
Measurement of Hip Circumference

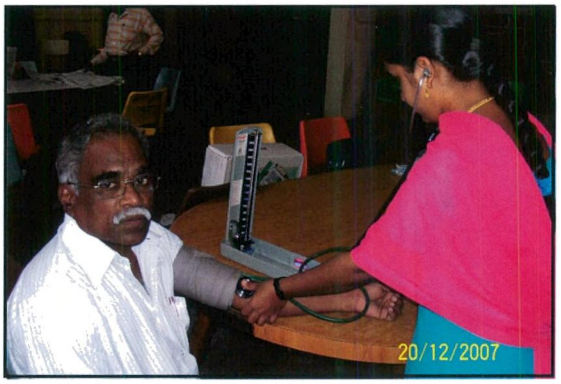

Plate 5
Measurement of Blood pressure

Plate 6
Drawing blood sample

The Blood Pressure of the adults was recorded with the help of a technician. Before taking the blood pressure reading, the adults were asked to be relaxed and comfortably seated, with their arm well supported. A cuff that inflates was wrapped around the upper arm of the adult and kept in place with Velcro, a fixing substance. Air was then blown into the cuff with increasing pressure until tightening was felt on the upper arm. The technician with the help of a stethoscope in the adults arm listens to the pulse while the air was slowly let out again. The systolic pressure was measured when the technician first heard the pulse. The diastolic pressure was measured from the moment the technician was unable to hear the sound of the pulse (Plate 5).

b. Clinical Examination of Symptoms

Physical examination or clinical examination is the process by which a health care provider investigates the body of a patient for signs of disease. It generally follows taking the medical history, an account of the symptoms as experienced by the patient. Together with the medical history, the physical examination aids in determining the correct diagnosis and devising the treatment plan (Reilly, 2003).

The selected hyperlipidemic and diabetic adults were screened and examined by a physician for the presence of clinical symptoms like giddiness, fatigue, shivering, polyuria, polydypsia, polyphagia etc. using a clinical assessment schedule (Appendix II).

3. Biochemical Assessments

Biochemical assessments are the most objective and sensitive measure of nutritional status. The adults were previously informed about the biochemical tests to be carried out the next morning and were asked to consume dinner by around 8 p.m. in the previous night. The biochemical tests for the adults were carried out after a 12 hours fasting i.e. around 8 a.m. in the next morning. The various biochemical tests done for the selected adults included blood haemoglobin, lipid profile, total cholesterol, triglycerides, HDL, LDL and VLDL

cholesterol. The lipid profile is a useful index for assessing the total cholesterol of an individual. The other biochemical parameters for diabetic adults included fasting blood sugar, post prandial blood sugar and glycosylated haemoglobin.

Collection of Blood Sample

The blood was collected from the selected adults by a well trained laboratory technician using disposable syringes. Five ml of blood was collected from each adult and 0.2 ml of the blood was used for the estimation of blood haemoglobin. The remaining blood sample collected from each adult was allowed to clot and the serum was separated according to the procedure obtained by Tietz (1990) and used for other biochemical estimations (Plate 6).

a. Blood Haemoglobin

Blood haemoglobin concentration is among the most commonly performed blood tests, usually as part of a complete blood count. Blood haemoglobin present in the blood sample is measured using the standard procedure given by Eilers (1967). Ferrous ions of the haemoglobins are oxidized to the ferric state by potassium ferric cyanide to form methaemoglobin. In turn, methaemoglobin reacts with the cyanide ions provided by potassium cyanide to form cyanmethaemoglobin, which has the absorbance at 540 nm.

The blood sample (0.2 ml) for the analysis was transferred into a tube containing the reagent. After five minutes of development of full colour, the optical density of the sample was measured at 530 to 550 nm against a reagent blank. The normal blood haemoglobin values for men range from 13.5 to 16.5 g / dl (Ashwood *et al.*, 1994).

b. Total Cholesterol

A total cholesterol test is a rough measure of all the cholesterol and triglycerides in the blood. Total cholesterol is an important measure of both bad and good cholesterol. In general, a total cholesterol value over 200 mg/dl

means a greater risk for heart disease. The total cholesterol values listed below were used as reference values (National Cholesterol Education Program, 2001).

Desirable : < 200 mg/dl

Borderline high : 200 to 239 mg/dl

High risk : > 239 mg/dl

The total cholesterol present in the blood sample was analysed using the Enzymatic colorimetric test CHOD – PAP method given by Allain *et al.* (1974). Cholesterol esters were hydrolysed to produce cholesterol. Hydrogen peroxide was then produced from oxidation of cholesterol by cholesterol oxidase. The indicator quinoneimine was formed from hydrogen peroxide and 4-amino antipyrine in the presence of phenol and peroxide. The absorption of the red quinoneimine dye was proportional to the concentration of the cholesterol in the sample. 10 µl of the serum was mixed, incubated for 10 minutes at 37°C. The optical density at 500 nm was measured within 60 minutes against a reagent blank.

$$\text{Cholesterol ester} + 2\,H_2O \xrightarrow{\text{CHE}} \text{Cholesterol} + \text{Fatty acid}$$

$$\text{Cholesterol} + O_2 \xrightarrow{\text{CHO}} \text{Cholesterol} - 3 - \text{one} + H_2O_2$$

$$2H_2O_2 + 4 - \text{Aminoantipyrine} + \text{Phenol} \xrightarrow{\text{POD}} \text{Quinoneimine} + 4\,H_2O$$

c. Triglycerides

Triglyceride level is used to measure the amount of triglycerides, a type of fat in the blood. A high triglyceride level may lead to atherosclerosis, which increases the risk of heart attack and stroke. Persons with high triglycerides often have other conditions such as diabetes and obesity that also increase the chances of developing heart disease and the following levels are used as reference values (National Cholesterol Education Program, 2001).

Normal	: < 150 mg/dl
Borderline High	: 150 -199 mg/dl
High	: 200 - 499 mg/dl
Very High	: > 499 mg/dl

Triglycerides present in the blood sample were analysed using the Enzymatic colorimetric test, GPO - PAP method given by Washfield *et al.*, (1975). Triglycerides were determined after enzymatic hydrolysis with lipases. The quinoneimine indicator was formed from hydrogen peroxide, 4 aminoantipyrine and 4-chlorophenol under the catalytic influence of peroxidase. 10 µl of the serum was mixed, incubated for 10 minutes at 37°C. The optical density was measured within 60 minutes against a reagent blank at 500 nm.

$$\text{Triglycerides} \xrightarrow{\text{LPL}} \text{Glycerol + Fatty acid}$$

$$\text{Glycerol + ATP} \xrightarrow{\text{GK}} \text{Glycerol-3-phosphate + ADP}$$

$$\text{Glycerol-3-phosphate + } O_2 \xrightarrow{\text{GPO}} \text{Dihydroxyaceton phosphate + } H_2O_2$$

$$2\ H_2O_2 + \text{Aminoantipyrine + 4-Chlorophenol} \xrightarrow{\text{POD}} \text{Quinoneimine + HCl + 4 } H_2O$$

d. HDL Cholesterol

HDL stands for High Density Lipoprotein, a form of "good" cholesterol. The laboratory test for HDL actually measures how much cholesterol is in the HDL, not the actual amount of HDL in the blood. HDL cholesterol was done using the Enzymatic method suggested by Friedwald *et al.,* (1972). The normal values for HDL cholesterol as given by National Cholesterol Education Program, (2001) are given below:

Low	: < 40 mg / dl
High	: > 60 mg / dl

e. LDL Cholesterol

LDL is a type of cholesterol and is usually done to determine the risk for heart disease. The LDL test is a part of lipid analysis, that measures the Low Density Lipoprotein (LDL) in the blood which also checks for total cholesterol, HDL and triglycerides. Too much LDL, commonly called "bad cholesterol," can lead to cardiovascular disease. According to the National Heart, Lung and Blood Institute, the lower the LDL, the lower the risk for heart disease or stroke. Too much LDL in the blood can clog arteries. A healthy LDL level is one that falls in the optimal or near-optimal range (NCEP, 2001).

Optimal	: < 100 mg/dl
Near Optimal	: 100-129 mg/dl
Borderline High	: 130-159 mg/dl
High	: 160-189 mg/dl
Very High	: > 189 mg/dl

LDL cholesterol was calculated using Friedwalds equation (Tietz, 1990)
LDL cholesterol = Total cholesterol – (HDL cholesterol + VLDL cholesterol)

f. VLDL Cholesterol

VLDL stands for Very Low Density Lipoprotein. VLDL contains the highest amount of triglycerides. VLDL is considered a type of bad cholesterol, because it helps cholesterol build up on the walls of arteries. Normal VLDL cholesterol level is between 5 and 40 mg/dl (NCEP, 2001). VLDL cholesterol was calculated using the formula given by Friedwald *et al.*, (1972).

VLDL cholesterol = Triglycerides / 5 where 5 is a constant factor.

g. Blood Sugar

Blood sugar can be estimated by the standard procedure of Enzymatic colorimetric test, GOD – PAP method given by Trinder (1969). Glucose was

determined after enzymatic oxidation in the presence of glucose oxidase. The Hydrogen peroxide formed reacts, under the catalysis of peroxidase, with 4 hydroxy benzoic acid and 4-aminophenazone to form a red violet quinoneimine dye as an indicator. 10 µl of the serum was mixed and incubated for 5 minutes at 37^{O}C. The absorbance of the sample and standard was measured against the reagent blank at 505 nm. The colour is stable for 30 minutes (Braham and Trinder, 1972).

$$\text{Glucose} + O_2 \xrightarrow{\text{GOD}} \text{Gluconic acid} + H_2O_2$$

$$2\ H_2O_2 + 4 - \text{Aminoantipyrine} + \text{Phenol} \xrightarrow{\text{POD}} \text{Chinonimine} + 4\ H_2O$$

Fasting Blood Sugar

The Fasting Blood Glucose (FBG) level is the most commonly used indicator of overall glucose homeostasis largely, because, disturbing events such as food intake are avoided. Abnormalities in these test results are due to problems in the multiple control mechanism of glucose regulation. Fasting blood sugar was estimated for all the selected diabetic adults. The desirable fasting blood sugar level is 80 to 115 mg / dl (Bamji *et al.*, 2003).

Postprandial Blood Sugar

The metabolic response to a carbohydrate challenge is conveniently assessed by a postprandial glucose level drawn one and half hours after a meal or a glucose load. The postprandial blood sugar levels of the selected adults were estimated one and half hours after the consumption of breakfast. The desirable postprandial blood sugar according to Bamji *et al.*, (2003) is 120 to 160 mg / dl.

h. Glycosylated Haemoglobin

Glycosylated (or glycated) haemoglobin (Hb_{1c}, or HbA_{1c}) is a form of haemoglobin used primarily to identify the average plasma glucose

concentration over prolonged periods of time. Long-term control of blood sugar concentration can be measured by the concentration of HbA_{1c} which is a product of the reversible reaction of haemoglobin A with glucose. A higher glucose concentration results in more HbA_{1c}. Because the reaction is slow, the HbA_{1c} proportion represents glucose level in blood averaged over the half-life of red blood cells, is typically 50 - 55 days (American Diabetes Association, 2007).

In the normal 120-day life span of the red blood cell, glucose molecules join haemoglobin, forming glycated haemoglobin. In individuals with poorly controlled diabetes, increases in the quantities of these glycated haemoglobins are noted. In general, the reference range found in healthy persons is about 4 – 5.9 per cent. The International Diabetes Federation and American College of Endocrinology recommend HbA_{1c} values below 6.5 per cent, while American Diabetes Association recommends that the HbA_{1c} be below 7.0 per cent for most patients. A high HbA_{1c} represents poor glucose control. Persistent elevations in blood sugar increase the risk for the long-term vascular complications of diabetes such as coronary disease, heart attack, stroke, heart failure, kidney failure, blindness, erectile dysfunction, neuropathy, gangrene and gastroparesis (slowed emptying of the stomach). Poor blood glucose control also increases the risk of short-term complications of surgery such as poor wound healing (Rohlfing *et al.*, 2002). The normal values of glycosylated haemoglobin as given by Trivedi *et al.,* (1978) are given below:

Non diabetic: 4 to 5.6 per cent
Good control: 5.6 to 7 per cent
Fair control: 7 to 8 per cent
Poor control: above 8 per cent

4. Food and Nutrient Intake

Food consumption pattern of the selected hyperlipidemic adults with or without diabetes was assessed using a three day 24 hour recall method.

Standard cups and measures were used during the recall method to help the adults to recall what they ate in the previous days. Food and nutrient intake was computed using the food composition table (Gopalan *et al.*, 2004) and compared with the Recommended Dietary Allowance (RDA) suggested for sedentary activity men (ICMR, 2004).

The dietary intake pattern including the details regarding the various foods restricted and avoided was also inferred using the schedule. All the adults were given diet counselling in order to ensure uniform pattern of food and nutrient consumption and equalization of dietary regimens in all the adults. Detailed instructions were given as to how to select the proper and correct amount of foods to adjust the energy content of the diet to keep up with the food items consumed. General instructions were given to the adults regarding the diet like avoiding roots and tubers, fats and oils, inclusion of more vegetables, green leafy vegetables and fiber rich foods.

D. Selection and Nutrient Analysis of Spices

1. Selection of Spices

From time immemorial, man has been extracting drugs from the herbs and spices. Because of general awareness of the widespread toxicity and harmful after - effects associated with the long term use of synthetic drugs for treating diabetes and other disease conditions, people have started treating their ailments by relying more firmly upon herbs and spices which are comparatively less exploited for their nutritive value and medicinal qualities (Bhattacharjee, 2000). After an extensive appraisal of literature pertaining to spices, cinnamon, cloves, garlic and turmeric were selected for the present study. These spices were chosen as they hold great scope for controlling blood cholesterol and blood sugar levels and are widely practised in Ayurveda system of medicine but lacks strong scientific evidence. These spices are commonly used in our Indian dietaries.

The spices selected for the study include

- Cinnamon *Cinnamomum zeylanicum*
- Cloves *Syzygium aromaticum*
- Garlic *Allium sativum* and
- Turmeric *Curcuma longa*

2. Processing of the Selected Spices

Preservation of foods by drying is one of the earliest and simplest techniques used for centuries. By definition, food dehydration is the process of removing water from food by circulating hot air through it, which prohibits the growth of enzymes and bacteria, without drastically reducing the taste and nutritive value of the foods. They keep well because of the combination of the physical changes. The principle of dehydration is that bacteria are unable to survive in the absence of moisture. This principle of dehydration has been used to process the spices for the present study.

The spices namely cinnamon, cloves, garlic and turmeric were procured from the departmental stores as a lot and cleaned to remove any impurities like stones, sticks, straws etc. Spices were washed with running water and allowed to dry under shade to remove the excess water. The cleaned spices were then spread in trays and dried in a cabinet drier at 40^{O} C for one hour. The trays were then removed and allowed to cool. The above process (i.e. heating at 40^{O} C for one hour and cooling) is repeated until the moisture content comes to less than 10 per cent. The dried spices were then pulverized using a pulveriser, sieved and then stored in air tight containers (Figure 3).

3. Nutrient Analysis of the Selected Spices

The nutrients present in the selected spices namely cinnamon, cloves, garlic and turmeric were analysed using the standard procedures given by NIN (1983). The nutrients analysed include moisture , energy, protein, fat, carbohydrate, ash and crude fibre; minerals include calcium, phosphorus,

sodium, potassium and iron; vitamins include thiamine, riboflavin, niacin, vitamin A and vitamin C; trace elements include lead, copper, arsenic, chromium and zinc; active principles include cinnamaldehyde, eugenol, allicin and curcumin.

E. Preparation of Capsules of Spice Powders and Food Costing

1. Preparation of Capsules of Spice Powders

Many research studies have suggested that two to five grams of herbal / spice powder can be consumed without any problem (Ken, 2006 and Dearlove, 2008). In the present study also, the investigator selected two grams each of the spice powder for supplementation. For long term treatment and effects, capsules and tablets are the most practical and convenient form.

The spice in the form of capsules was thought to be more acceptable than given as such due to its strong flavour and taste. Hence it was thought of interest to prepare capsules for supplementation. Many of the adults of the present study preferred the spices in the form of capsules as revealed from the interview, the investigator prepared the spices in the form of capsules with the help of manufacturing company. The capsules were filled with approximately 490 to 500 mg of spice powders to facilitate easy distribution of appropriate dosage (Figure 1).

2. Food Costing

The cost of capsules prepared using cinnamon, cloves, garlic and turmeric have been computed and compared with the allopathic drugs which are used for controlling hyperlipidemia and diabetes mellitus available at the standard pharmacies.

The details of the supplementation study were presented before an Ethical Committee constituted by the University and approved by the Committee members (HEC.2007.08).

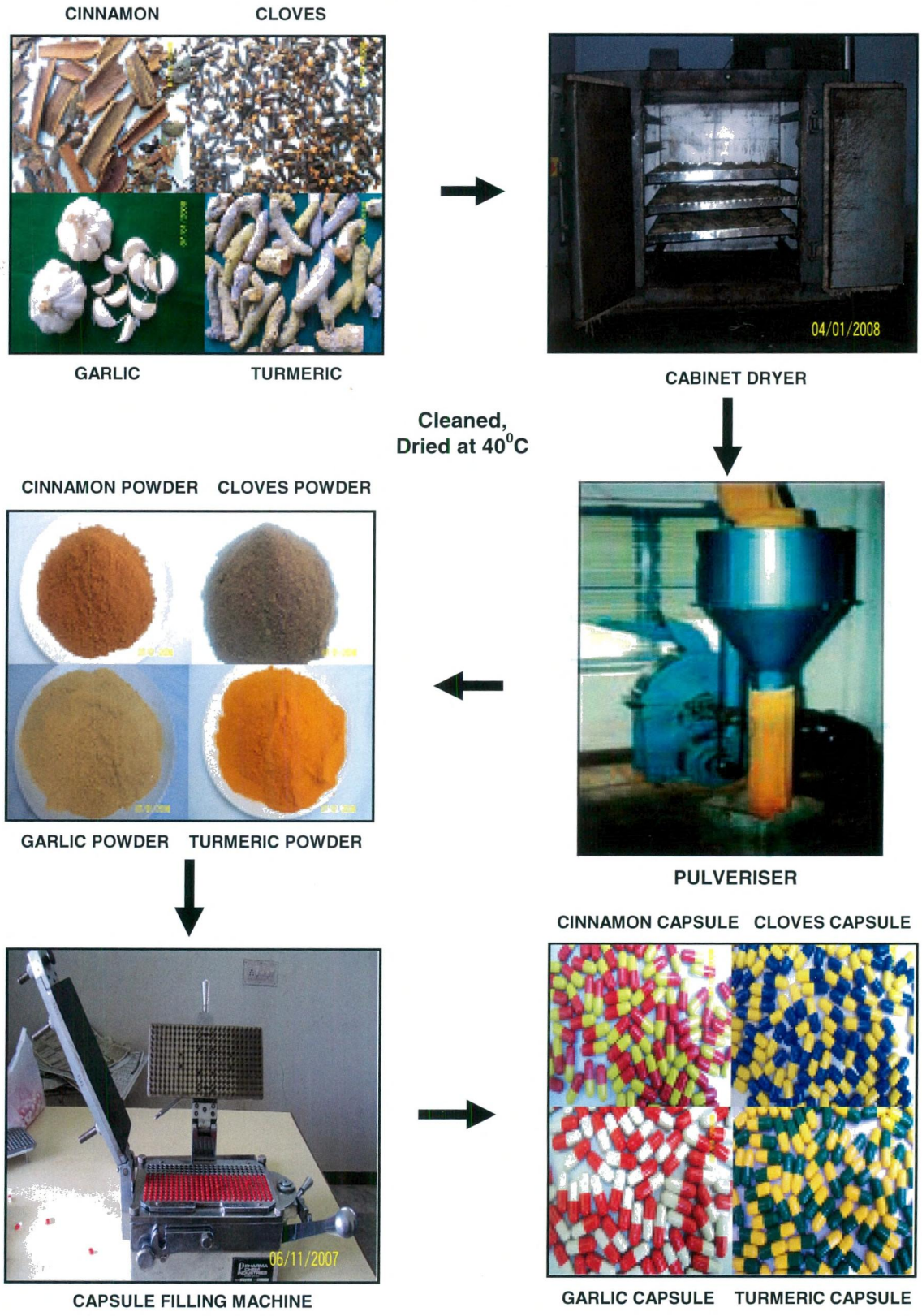

FIGURE 1

PROCESSING OF SPICES INTO CAPSULES

PHASE II

F. Conduct of the Supplementation Study

For the conduct of the supplementation study, hyperlipidemic adults were selected from Lakshmi Machine Works Limited, Periyanaickenpalayam and the adults with hyperlipidemia and diabetes mellitus were selected from BSNL, Bharathi Park Road, Coimbatore. These concerns were selected because more adults will be available from one area, which is convenient for the investigator to give supplements and take blood samples for analysis and monitor the study. Prior permission was obtained from the concerned authorities for conducting the study.

The importance of spices and their medicinal value such as lipid lowering effects and sugar controlling effects have been explained to the adults selected for the study. It was made sure to the adults that these spice capsules were only food supplements and will not cause any side or harmful effects on their health.

The selected 75 hyperlipidemic and 75 hyperlipidemic with diabetic adults were divided into five groups consisting of 15 adults in each group. Sixty adults (four groups of 15 adults in each group) constituted the experimental group who were given each two grams of cinnamon, cloves, garlic and turmeric in the form of capsules daily for a period of three months. The remaining 15 adults constituted the control group. To the hyperlipidemic control group, two grams of powdered sugar and to the diabetic control group two grams of roasted bengal gram flour in the form of capsules were given daily till the end of the supplementation period.

The adults were asked to consume four capsules per day i.e. two capsules after breakfast and two after dinner for a period of three months.

The investigator visited the selected adults at their places fortnightly and gave the supplements in the form of capsules. The investigator also ensured that the adults in the supplementation groups consumed these supplements regularly through regular visits and telephonic random enquiry. The adults were asked to report to the investigator immediately if they came across any disturbance or inconvenience throughout the supplementation period while consuming these spice capsules.

G. Evaluation of the Supplementation Study

The effect of supplementation on the selected adults was evaluated using the anthropometric measurements like height, weight, BMI, WHR and blood pressure; clinical assessment for the presence of symptoms; and biochemical assessment including blood haemoglobin, total cholesterol, triglycerides, HDL cholesterol, LDL cholesterol, VLDL cholesterol, fasting blood sugar, postprandial blood sugar and glycosylated haemoglobin before and after a period of three months of supplementation. Feedback was obtained from all the selected adults at the end of the supplementation study.

The overall research design is presented in Figure 2 and the supplementation study is presented in Figure 3.

H. Analysis of the Data

The data collected were consolidated and statistically analysed for arriving at the results of the effect of supplementation of the selected spices on hyperlipidemic and diabetic adults for various parameters.

PHASE I

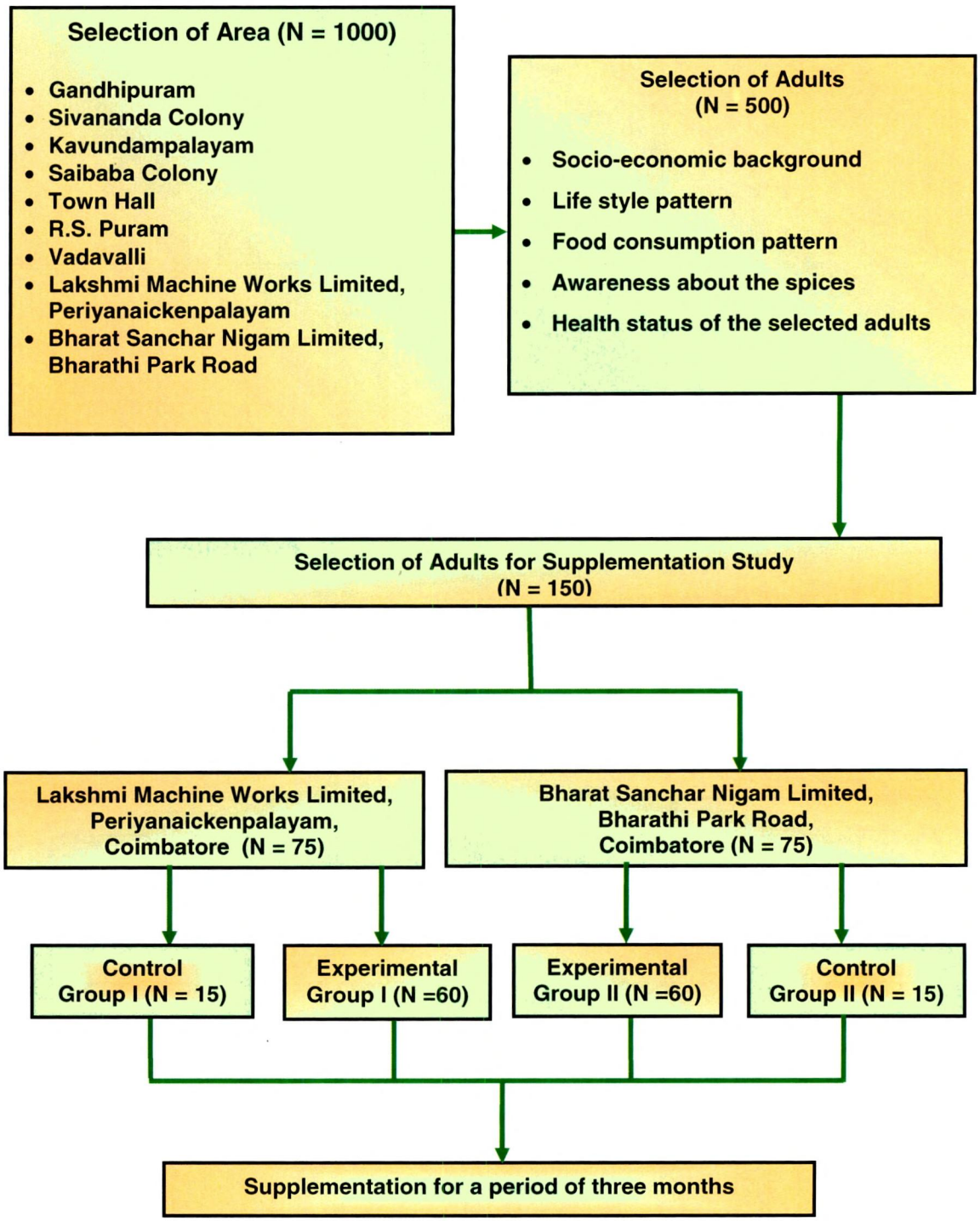

FIGURE 2

RESEARCH DESIGN

PHASE II

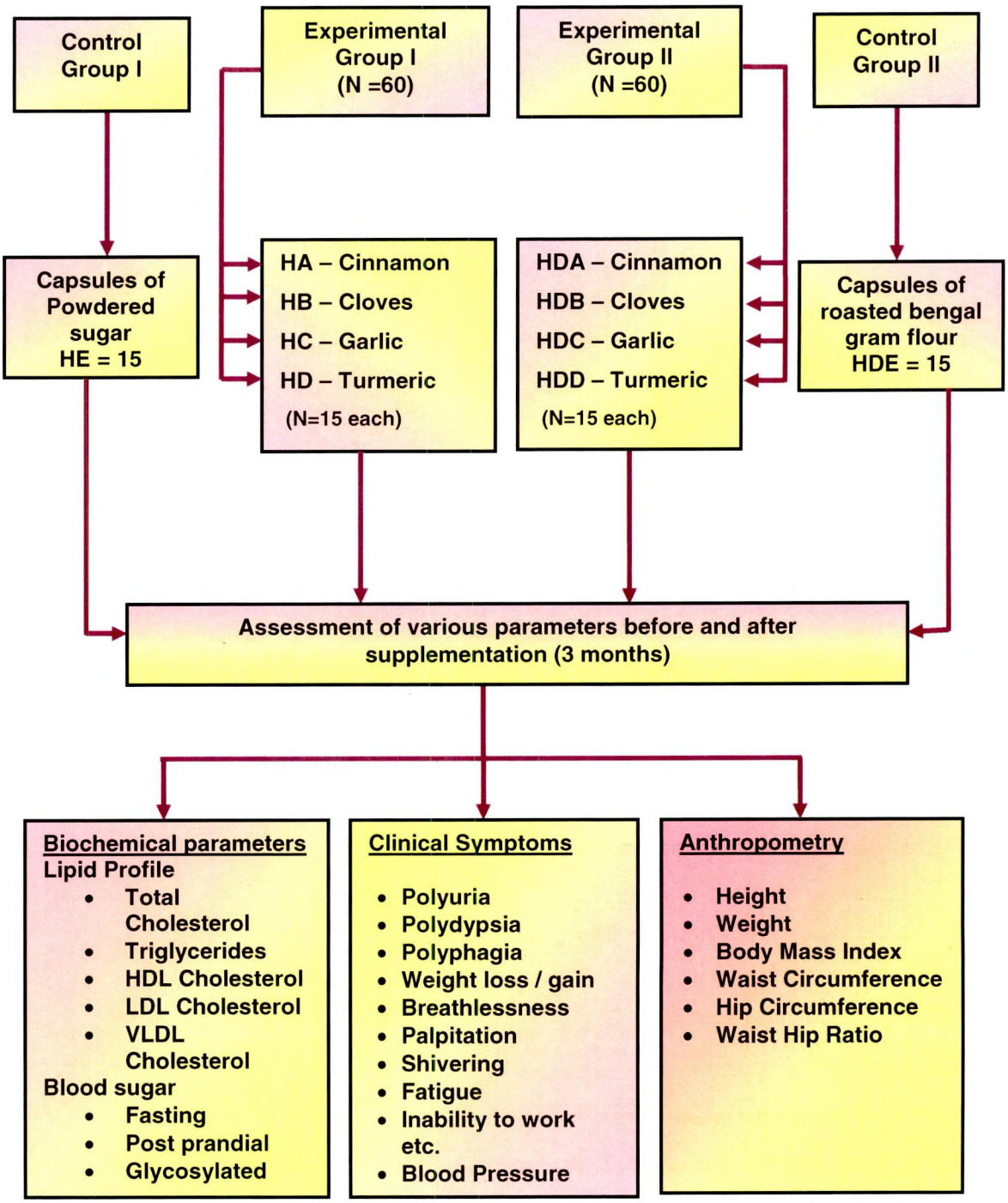

FIGURE 3

SUPPLEMENTATION STUDY