Biochemical Profile of Selected Normal Healthy Adult Men and Women

BY

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Introduction

I. INTRODUCTION

"Let us communicate health rather than disease. Let's talk about it! Nothing is possible without health - Let's talk health", was the slogan given by Hiroshi Nakajima (1989) in the World Health Assembly.

The World Health Organisation defines health as "a state of complete physical, mental and social well being and not merely the absence of disease or infirmity". Winter (1989) explains that the health is the science and art of helping people change their life style to move toward a state of optimal health. Optimal health is defined as a balance of physical, emotional, social, spiritual and intellectual health.

The healthy person is more optimistic, courageous and happy. Good physical health promotes favourable attitude and favourable attitude vitalises the body's tissues and organs, resulting in maximum efficiency.

According to Mahler (1982) health can only be attained through a combination of diverse measures. Such health measures are the cornerstone of the strategy for attaining "health for all" by the year 2000 A.D.

Accordingly, every man, woman and child should be in a position to choose a healthy way of life. To do this, they must be adequately informed on matters that have an influence on health, the environment, water, food, good habits and bad ones.

Kuku (1988) states that there still exists a gap between the health status of the people in the developed and developing countries and even among the people within a country. Nearly 1000 million people are trapped in the vicious circle of poverty, malnutrition, disease and despair that saps their energy, reduces their work capacity, and limits their ability to plan for the future (Mahler, 1988). The nutritive of the individual affects productivity to a very great extent. A well nourished adult is said to have a good working capacity and increased productivity. Nutritional well being is a pre-requisite for health, full functional capacity and physical fitness of individuals and populations (WHO, 1988).

According to Kumar and Kumar (1988), "Out of 500 million women in developing countries (excluding China) about 70 million are pregnant at a point of time and nearly two thirds of them i.e. 48 million have anaemia.

Women, world over are becoming increasingly aware of health and healthy life styles. Modern women now realise that the health of their husbands and children is a product of food and the life styles they themselves create. There are approximately 23.9 million women in Tamil Nadu and they are capable of influencing the health of their families and society at large. According to Krishnaswamy (1989) a majority of them are as yet unaware of the importance of their own health.

Shryock (1987) exhorts that a person must pay for his good health by willingly forfeiting personal indulgences that substract a person's general vitality. He must train his appetite to enjoy the kinds of food that serve his body best in providing energy and in furnishing the food elements needed for tissue building and repair. The health conscious person must choose not to smoke and he abstains from alcoholic drinks. A healthy person is more efficient than one who is not healthy .

As a signatory to the Alma Ata Declaration (1978) India has rightly placed in its National Health Policy greater stress on health care than on medical care.

Developmental programmes place emphasis on the improvement of nutritional status of people as the principal goal. The emphasis is clearly on the preventive and promotive aspects of health. This implies that the responsibility for the health of the people must lie primarily with the people. It may be realistic to proceed on the basis that the health of the nation lies in the hands of the people themselves.

It is an acknowledged fact, according to Williams (1985) that the productivity is a basic requisite for obtaining the economic, social and political goals of the country and for raising the standard of living of the people to enable the country to join the cadres of developed countries. Hence, it is essential for any government or nation to ensure that their population is helped to achieve health and optimum nutritional status which is the foundation for socio-economic development.

Adequate data has been collected by developed and developing nations with reference to anthropometric measurements and dietary data for the assessment of the nutritional status of adults, taking into consideration

the ecological factors prevailing in these developed countries (ICMR, 1984). However, such organised data is not available for our country with special reference to bio-chemical profile, where there is a wide variation among the environmental, socio-economical, racial and cultural factors.

Garn et al (1976) found a racial difference in haemoglobin concentrations, with median values for blacks of all ages about 1g/dl below those for whites. The difference could not be explained by differences in socio-economic status or geographic location. Such findings suggest the need for race specific,national and region specific standards for evaluating biochemical data.

Busina (1981) stresses that there is obvious need out of public health importance to study the health implications of biochemical parameters of nutritional status and for developing standards or guidelines which are derived from experimental and clinical experience. In this context, it would be important to have more information about the levels of biochemical parameters

which could throw adequate light on health problems which could be prevented in the early stages.

Varley et al (1980) suggest that it has long been customary to quote "normal ranges" for the concentration of constituents in body fluids to indicate the values found in healthy persons.

A beginning was made by the National Institute of Nutrition towards providing guidelines for anthropometric measurement of healthy groups and dietary intake of adults. But little progress had been made towards developing biochemical profile of healthy adults in developing countries specially in India. Data along these lines are important if a country wants to have a realistic picture of the health status of its population.

Hence, the present study was undertaken with the following objectives:

1. To select healthy adult men and women and assess their food and nutrient intake and collect information on their past health history.

2. To determine the biochemical profile of these healthy adult men and women with regard to selected nutrients.

3. Develop data which can be used as reference material for our population groups.

Review of Literature

II. REVIEW OF LITERATURE

The review of literature pertaining to the study on the "Biochemical profile of selected normal healthy adult men and women", is based on the following headings:

1. Need for Assessment of Nutritional Status of individuals.
2. Need for developing local norms for biochemical Profile.
3. Biochemical profile of individuals with regard to the following nutrients:
   a. Haemoglobin, Packed Cell Volume and Mean Corpuscular haemoglobin content.
   b. Serum iron, total iron binding capacity, unsaturated iron binding capacity and percentage saturation of transferrin.
   c. Serum total protein, albumin and globulin.
   d. Serum retinol
   e. Serum calcium
   f. Serum zinc and copper
   g. Blood Glucose, and
   h. Serum cholesterol.

1. Need for Assessment of Nutritional Status of Individuals

According to Greaves and Berry (1978), nutritional Status is that condition of health of people that can be attributed to the foods they habitually consume. Nutritional assessment is the process whereby the state of nutritional health of an individual, or group of individuals is determined (Robinson et al, 1982).

Jelliffe (1978) points out that the principal aim of the nutritional assessment of a community is to map out the magnitudes and geographical distribution of malnutrition as a public health problem, to discover and analyse the ecological factors that are directly or indirectly responsible and where possible suggest appropriate corrective measures preferably capable of being applied with continuing community participation.

According to Jelliffe (1966), Assessment of Nutritional Status is necessary to define human nutritional problems and to guide the practical efforts towards their solution.

Nutritional status refers to the health of an individual as influenced by the intake and utilization of nutrients. It includes anthropometric, clinical, biochemical and dietary data. The conclusions reached through nutritional assessment become the basis of intervention programmes in the community and for the planning and implementation of nutritional care of individuals. Dietary counseling is based upon the information obtained by nutritional assessment and entails, planning, implementation and evaluation (Robinson et al, 1982).

Nutrition indicators can be popularly more acceptable to use than economic measures of poverty and are closely correlated with other indices of social and economic status (Thompson et al, 1982).

Bjorn Isaksson (1974) point out that in determining the nutritional status of an individual, the methods used involve a careful diet history, a physical examination and relevant laboratory tests.

The gradual tissue desaturation of nutrients and biochemical lesions that precede the anatomical lesions and signs of deficiency diseases are shown diagramatically in Figure 1 by Krehl and Hodges (1974).

Diagramatic Representation of occurrence of biochemical lesions

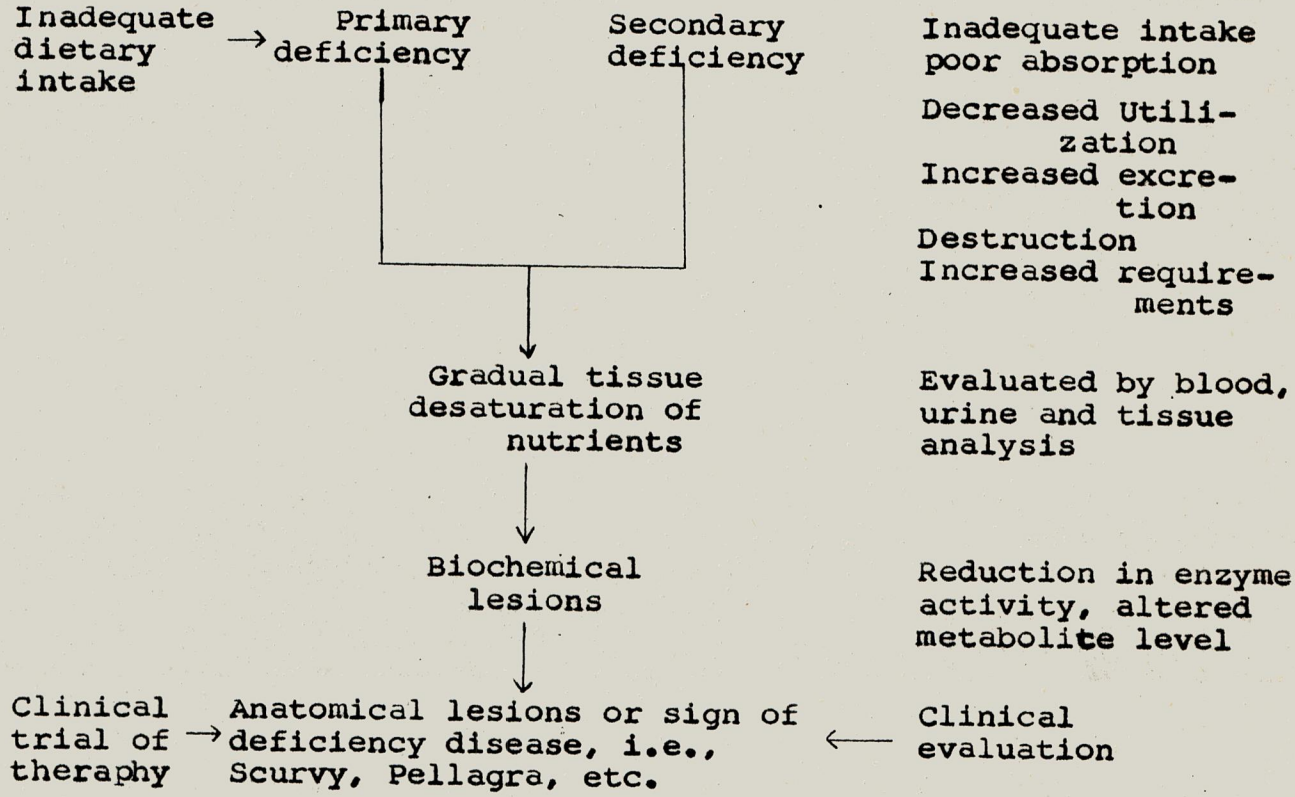

Figure - 1

Anthropometry deals with comparitive measurements of the body. According to Jelliffe (1978), nutritional anthropometry is concerned with the measurement of the variations of the physical dimensions and the gross composition of the human body at different age levels and degrees of nutrition.

Burk (1978) stressed that anthropometric measurements provide indications of the physical growth and development of individuals. The most frequently used are height and weight, often supplemented by arm circumference and triceps skinfold.

Devadas (1986) opines that although there are many variables related to growth, the most commonly and widely used single measurement is the body weight of an individual.

Weisell and Francois (1982) reveal that anthropometric measurements provide basic information used in assessing the nutritional status, determining protein and energy requirements and reviewing food supplies.

Gray and Gray (1980) also stress that anthropometry provides atleast one quick and easy way to assess the patients protein and calorie reserves.

According to Beaton and Bengoa (1982), height remains the standby for measurement of skeletal growth. It probably correlates better with socio economic status in most age groups than soft tissue measures or weight.

The current consensus according to Chowdhury et al (1985) is that the ultimate value of an anthropometric index rests on its capacity to discriminate and identify individuals and populations at high risk, of the functional consequences of malnutrition-mortality and morbidity.

Jelliffe (1979) opines that optimal dietary information provides enough information to quantity individual calorie, protein and vitamin intakes for the period corresponding to the survey or study.

According to Garn et al (1975), a good 7-day intake information, covering the period immediately before the anthropometric, physical and biochemical examination is surely to be desired, certainly optimal.

Thimmayamma (1987) opines that among several methods of diet survey, weighment method has been followed by several workers for assessing the family dietary intake.

According to Bamji (1983), diet surveys can help to detect inadequate intake of nutrients. However, they give no utilization defects. Clinical examination would fail to identify people at risk, unless sensitive tests for detecting functional impairments of the type mentioned above can be defined. Biochemical tests are the only means of detecting subclinical nutrition. Further they help to establish the clinical diagnosis.

An ideal biochemical test should be specific, minimally invasive (Performable on blood or urine) and preferably simple and inexpensive. (Bamji, 1983).

Osancova (1979) observes that biochemical methods are used for correlation with clinical examinations and are as a rule able to indicate deficiencies sooner than actual clinical symptoms develop.

2. Need for developing local norms for biochemical profile

Just as malnutrition is a composite problem of man, his food and his environment, so is nutritional status. India along with many other countries has a young population. The income levels are low and about half the population live below the poverty line. Often the foods suggested are beyond the reach of the community (Ghosh, 1976). The availability of food affects the resulting health which is central to the study of nutrition. Particularly striking examples of this fact can be seen in the classic symptoms associated with some vitamin deficiencies (Mullen et al, 1984). Apart from these, a number of ecological factors like food availability and food production, environmental factors, socio-economic, racial, climatic and cultural variations influence the nutritional status of individuals and may alter the biochemical profile (Williams, 1985).

A standard embodies, the concept of a norm or target - that is a value judgement in the above mentioned circumstances and in which local standards are preferred (WHO, 1986).

Busina (1981) stresses that there is obvious need out of public health importance to study the health implications of biochemical parameters of nutritional status and for developing standards or guidelines which are derived from experimental and clinical experiences. In this context, it would be important to have more information about the levels of biochemical parameters which could throw adeduate light on health problems which could be prevented in the early stages.

In the words of Mertz et al (1980), biochemical tests of metabolites or nutrients in the blood is a good indicator of nutritional status and setting up of local norms pertaining to the various nutrients in it, forms the basis of the study.

3. Biochemical Profile of Individuals with regard to the following nutrients

   a. Haemoglobin, Packed Cell Volume and mean Corpuscular haemoglobin content

Anaemia is a broad term applied to the condition in which there is inadequate or defective formulation of haemoglobin and defective saturation and formation of red blood cells (Swaminathan, 1985).

According to Johnson (1984), the major laboratory tests used in the diagnosis of anaemia and iron deficiency include the analysis of haemoglobin, mean corpuscular haemoglobin content, and total iron binding capacity. Values for all these are known to vary to some degree according to age and or sex (Bothwell et al, 1980).

Cook et al (1977) stresses that a far more accurate index of iron nutrition can be obtained by using haemoglobin concentration in conjunction with the transferrin saturation, serum ferritin, and free erythrocytic porphyrin.

According to Garn (1979), the haemoglobin and hematocrit norms used should be derived from adequately large samples and carefully age-adjusted.

Kritchevsky (1980) exhorts that functional iron is most commonly assessed by the determination of haemoglobin concentration and of Packed Cell Volume (PCV, hematocrit) and by the calculation of red cell indices such as mean corpuscular haemoglobin content.

According to Hereburg and Galan (1985), the absorption of dietary iron reflects the iron status of the individuals. The significant biochemical indicators of iron status are very sensitive to any external or internal health problems.

According to Dallman (1986) the concentration of haemoglobin provides the best frame of reference for severity of iron deficiency both in rat and in man because it is the most easily measured essential iron compound.

Ernst et al (1986) opines that PCV is one of the influencing factors of blood rheology.

Vijayalakshmi et al (1987) suggested mean and range value of haemoglobin and Packed Cell Volume for adult men, in the age group of 25-30 years, as 13.9g/dl and 12.0 - 14.0g/dl and 39.75 per cent and 37-44 per cent, respectively.

b. Serum iron, total iron binding capacity, unsaturated iron binding capacity, percent saturation of transferrin

Iron deficiency is the most common nutritional disorder in the world, both in developing and developed countries. It is also the most common cause of nutritional anaemia. When widely prevalent and severe in magnitude, it has detectable and significant effects on physical performance, resistance to infectious diseases, and behavioural and cognitive functions (Tanphaichitr, 1983).

The laboratory measurements of serum transferrin is a valuable adjunct in the assessment of both iron and protein nutritional status (Mohammed and Gundi, 1988).

Taylor et al (1989) stress that although both serum ferritin and transferrin saturation reflect iron status in iron depleted subjects, the control of iron absorption in iron replete subjects is more dependent on iron stores as reflected in the serum ferritin concentration than on the percent saturation of transferrin.

Available data indicates clearly that in all age group studies, physical performance and productivity are

adversely affected by anaemia and that iron supplementation of anaemic workers improves their physical performance as well as productivity (Gopaldas et al, 1988).

National Institute of Nutrition (1983) reports that women in all centres studied, manifested higher prevalence of anaemia than men (50 per cent and 70 per cent in women Vs 30 per cent - 50 per cent in men).

Seshadri et al (1984) stress that anaemia clearly reduces physical performance and productivity.

Basta et al (1979) report that studies on adult men from Indonesia have demonstrated a significant difference in productivity in non-anaemic (Hb 13g/dl) Vs anaemic (Hb 13g/dl) tappers and weeders. Non-anaemic tappers collected 18.7 per cent more latex than anaemic tappers. Iron supplementation of the anaemic workers increased productivity.

Battacharya (1984) quoted the normal values of serum iron and total iron binding capacity as 50-175mcg/dl and 300-360 mcg/dl for both men and women respectively.

According to Rebecca (1982) and Cook et al (1982), serum iron level, transferrin saturation co-efficient and erythrocyte protoporphyrin concentration are useful in indicating the adequacy of the iron supply to erythroid marrow.

C. Serum Total Protein, Albumin And Globulin

The use of serum protein level as markers of nutritional status relies on the relationship between hepatic synthesis of substances and substantiate availability which is presumably related to nutritional status (Golden et al, 1982).

According to Young et al (1988), assessment of the status of the dynamic state of amino acid and protein metabolism in man is worth while in relation to the study of the normal metabolic changes taking place during growth and development, as well as for understanding the changes in metabolism that occur when the level and/or balance or amino acid intake is altered.

Serum Protein levels are significantly decreased in all categories of PCM. A corresponding significant

decrease in serum albumin was also seen in all categories which was due to decreased synthesis in the liver (Sharma et al, 1981).

Blackburn (1980) suggests that visual protein sources can fall while somatic protein sources remain within normal range. Some evidence, however, suggests that, in certain cases, plasma albumin may drop and kwashiorkor may develop before the anthropometric measures indicate depletion.

Watson et al (1985) opine that PCM is a major contributing favor facilitating infection in malnourished populations in the developing areas of the world. Reduced concentrations of Immunoglobulin would enhance the ability of mucosal pathogens to attach to epithelial surfaces and replicate.

According to Cornn and Cornn (1980), the range of acceptable levels with reference to total serum protein, albumin and globulin for men and women are 6.0 - 8.0/dl, 3.5 - 5.5g/dl and 2.5 - 5.5.g/dl respectively.

Khan and Chakraborti (1986) observe that diet low in iron and adequate in protein resulted in an increase in total plasma proteins.

Serum albumin decreased with age in women but not in men (Watler et al, 1985).

d. Serum Retinol

According to Swaminathan (1985), inadequate intake of Vitamin A or pro Vitamin A (active carotenoids) results in a decrease in the levels of Vitamin A in blood. The ICNND Mannal for Nutrition Surveys indicates that for young adults serum Vitamin A levels between 20 to 50 mcg/dl is normal, 10 to 20 mcg/dl is sub normal and less than 10 mcg/dl is deficient.

Serum Vitamin A measurements remain the only practical biochemical means of assessing the nutritional status of this nutrient. Prolonged low intakes of Vitamin A correlate with serum Vitamin A levels (Sauberlich, 1976).

Hodges et al (1978) found that haemoglobin values decreased in a pattern similar to that of plasma Vitamin A and that during repletion with Vitamin A haemoglobin values increased along with plasma Vitamin A.

According to Bendich et al (1989), in humans, most obvious and clinically important manifestation of Vitamin A deficiency is the eye disease xerophthalmia, which can lead to permanent blindness.

Sommer (1982) took serum albumin as marker of protein status and found that it was inversely related to the severity of xerophthalmia.

WHO (1987) observe that deficiencies caused by insufficient Vitamin A intake or impaired absorption strikes most dramatically at the eyes, producing night blindness, keratinization of the conjunctiva and cornea, corneal ulcertion and necrosis (death) of the cornea.

According to Bauernfeind (1980) the early stages of xerophthalmia including the characteristic symptom of night blindness are reversible but the condition becomes irreversible upon ulceration of the eye tissue.

Olson (1986, 1987) and Taylor (1986) caution that serum retinol levels are closely controlled and do not increase with increasing dietary intake except in deficiency status.

National Health and Nutrition Examination Surveys (NHANES, USA, 1987) panel suggests that serum Vitamin A data (retinol) could provide some indication of Vitamin A nutritional status in population groups.

Causal relations of malnutrition and Vitamin A deficiency

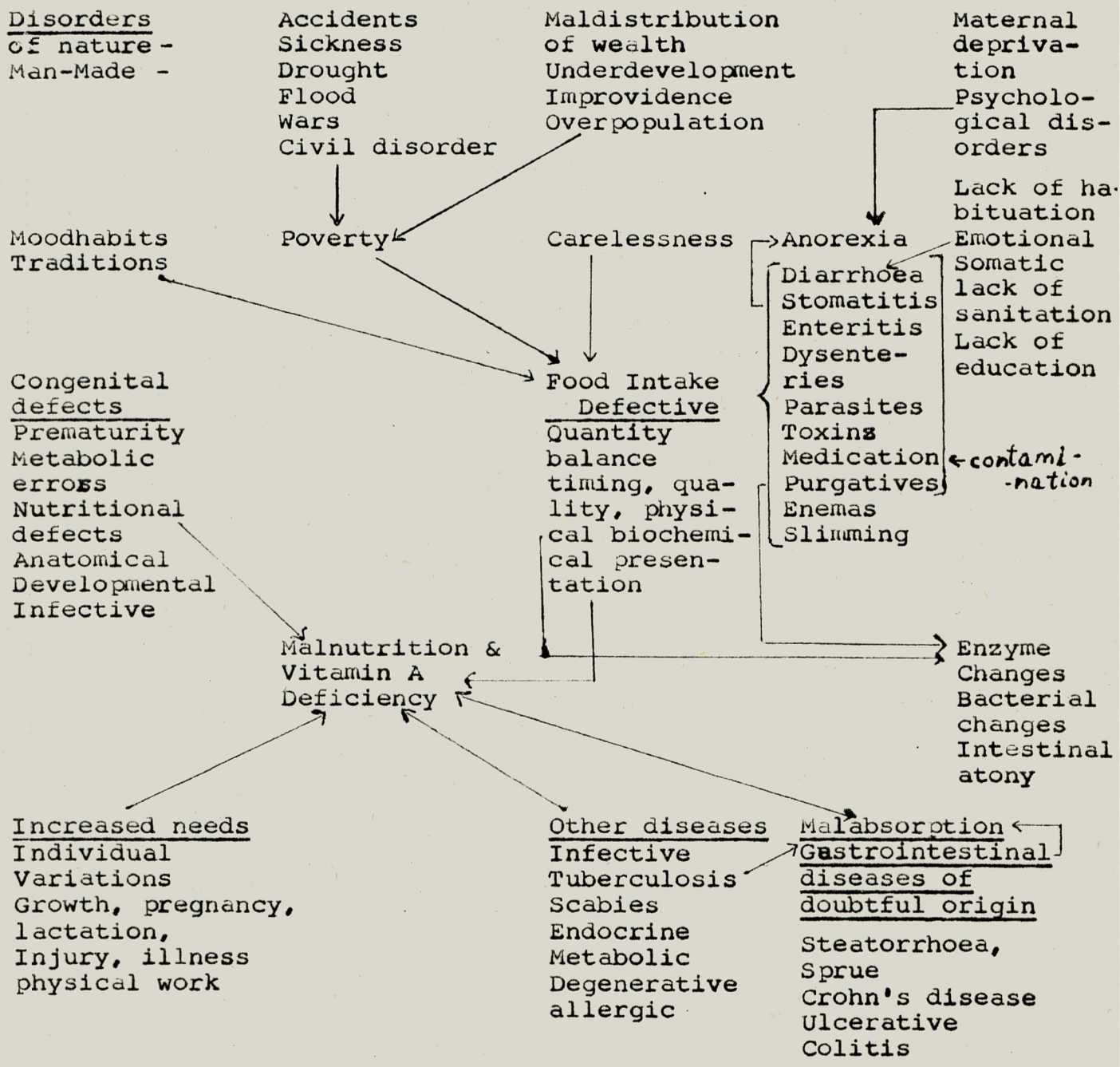

Figure - 2

e. Serum Calcium

According to Sauberlich (1976), the normal range for human serum calcium is quite narrow, probably about 9 to 11mg/dl. Blood calcium is controlled so closely that should it vary outside the normal range, one might suspect pathological problems before nutritional aspects.

McCarron (1987) observes that marginal intake of calcium raises blood pressure in normal rate.

According to Johnson et al (1985) hypertensive people excrete more calcium than normotensives and respond differently to calcium supplementation; urinary calcium is unchanged in the former and increased in the latter.

Sowers et al (1985) report that calcium supplementation was found to lower diastolic blood pressure of healthy Panamian young adults, with a greater effect noted for male than female subjects.

According to Altura and Altura (1985), increased intracellular calcium concentrations are the putative source of vaso constriction in the microcirculation.

According to Swaminathan (1981), of the total calcium of 6-11mg/dl,60 per cent is present as ionised calcium (5.4-6.6mg). This fraction has a great clinical importance. A decrease in the ionised calcium content below 3 mg will lead to the development of tetany and an increase in ionised calcium content to over long may cause renal failure and cardiac arrest.

f. Serum Zinc and Copper

Smith et al (1980) and Fisher et al (1981) through a number of experiments have demonstrated that copper absorption is diminished when dietary levels of Zinc are high.

Owens (1982), and National Centre for Health Statistics (1983), report that plasma Copper is higher in blacks than in whites; both plasma copper and zinc have been shown to decrease with increasing age.

Sloane et al (1985), observe that induced copper deficiency in calves first reduced serum copper and secondly slowed growth.

Greger et al (1978), report that dietary intakes of copper and zinc were not positively related to plasma levels of the elements.

Gentile et al (1981), demonstrated a significant positive correlation between hair zinc concentration and age, height and weight in 49 healthy subjects.

According to Haynes et al (1985) inadequate levels of dietary zinc have profound influence on growth, development and immune function in human populations. Marginal zinc deficiency in man is now believed to occur throughout the world including developed countries such as the USA (Hamb idge et al, 1976).

Peterson and Bettger (1986) found that after one week of zinc deprivation hematocrit values increased significantly.

The range of acceptable levels of serum zinc and copper, suggested by Vijayalakshmi et al (1984) for adult men are (25 - 35 years) 74 -114 mcg/dl and 78 -160mcg/dl respectively.

g. Blood Glucose

According to Levine (1986), in healthy persons glucose homeostasis maintains blood glucose levels between 70 and 130 mg/dl, despite perturbation by meals, fasting and exercise.

Studies by Dulloo et al (1985) have indicated that prolonged consumption of high sugar diets leads to increases in blood levels of cholesterol, triglycerides, fasting insulin and alters platelet behaviour.

Guar gum can reduce post prandial blood glucose insulin requirement and serum total cholesterol levels in Type I diabetic patients (Periti et al, 1988).

Glucose has been reported to enhance the intestianl absorption of calcium in humans (Normaň et al, 1980) and zinc in humans (Steinhardt et al, 1984).

Sud et al (1988) opine that Nutrient composition is a poor predictor of glycaemic response.

Krishnamachar and Mickleson (1987) have pointed out that individuals in a high state of physical training have lower blood glucose levels than sedentary individuals following the ingestion of a carbohydrate load.

h. Serum Cholesterol

Cholesterol concentration in the plasma has been shown to be an independent predictor risk of developing ischaemic heart disease (Lukaski et al, 1984).

Hartung et al (1980) found that total cholesterol decreased and high density lipoprotein cholesterol (HDL-C) increased directly with the amount of physical activity.

Masarai et al (1984) found serum total cholesterol levels to be lower at all ages in male and female seventh-day adventist vegetarians and triglyceride levels to be higher in men over 35 years of age.

Miller et al (1985) observe that cholesterol in the egg increases blood cholesterol concentration which leads to deposition of the sterol in the coronary arteries and promotes atherosclerotic heart diseases.

Phillipson et al (1985) reported that lipemic (Type IIb and Type V) subjects consuming fish oil rather than vegetable oil or a mixture of dietary fats evidenced a significant fall in serum cholesterol and triglyceride levels.

Sidhu and Dakenfull (1986) present that control of plasma cholesterol and nutrient absorption through dietary sapomins could provide substantial health and nutritional benefits in humans.

The range of acceptable level of serum cholesterol for both adult men and women is 150 - 240 mg/dl.

Methodology

III. METHODOLOGY

The methodology relating to the study on the "Biochemical Profile of normal healthy Adult men and Women" is framed along the following lines:

I. Selection of Venue

II. Selection of Volunteers

III. Assessing Nutritional Status of the Volunteers

1. Recording Anthropometric Measurements
2. Recording the Health Status through Clinical Examination
3. Recording Food and Nutrient Intake
4. Recording Blood Pressure and Pulse Rate.

IV. Determination of Nutrient levels in Serum

A. Collection of Blood and Separation of Serum

1. Estimation of Serum Iron and Total Iron Binding Capacity
2. Estimation of Total Serum Protein and its Fractions Albumin and Globulin
3. Estimation of Serum Retinol
4. Estimation of Serum Calcium
5. Estimation of Serum Zinc and Copper
6. Estimation of Serum Cholesterol

B. Analysis of Blood

1. Estimation of Haemoglobin
2. Estimation of Blood Glucose

V. Analysis of Data.

I. Selection of Venue

For this study, Tamil Nadu Agricultural University and Avinashilingam Institute for Home Science and Higher Education for Women (Deemed University) in Coimbatore were selected as the venue. The availability of young healthy adults was the main reason for choosing these places as venue for the study. Since all the volunteers were educated they could understand the purpose of the study and hence were very cooperative.

II. Selection of Volunteers

One hundred and one volunteers were selected for this study having the following as guidelines for selection:

1. Volunteers from both male and female sexes who were willing to participate and cooperate for the present study were chosen as the subjects.

2. There were on the whole, 50 adult men and 51 adult women volunteers participating in the study. All the volunteers were in the age group of 20-39 years of age as per the guidelines suggested for a Reference Man and Reference Woman by the Indian Council of Medical Re search (1984).

3. The selected volunteers were physically fit for active work and were usually engaged in moderate activity.

4. All the volunteers consumed a balanced diet and were not undergoing any diet restriction or drug treatment.

5. The selected volunteers had not suffered any major illness or accident in the immediate past two years.

6. The volunteers were free from hereditory disorders like diabetes and others.

7. The volunteers chosen were non-smokers, non-alcoholics and enjoyed good health. The alcoholics were avoided because, according to Walker et al (1980), Robinson et al (1982), Harvey et al (1985) and Artidevan (1986), despite good nutrition, long term alcohol consumption produces undesirable changes in the normal biochemical profile of an individual.

III. Assessing Nutritional Status of Volunteers

1. Recording Anthropometric Measurements

According to Gray and Gray (1980), anthropometry provides atleast one quick and easy way to assess the patient's protein and energy reserves.

Weisell et al (1982) proposes that anthropometric data provides basic information used in assessing the nutritional status, determining protein and energy requirements and reviewing countries food production and supplies.

Bairagi et al (1985) observe that the current consensus is that the ultimate value of an anthropometric index rests on its capacity to discriminate and identify individuals and populations at high risk and alert them about the functional consequences of malnutrition - morbidity and mortality.

In the present study the volunteers were weighed on a beam type scale without shoes and with the same light garments each time following precautions and guidelines suggested by Jelliffe (1966). Swaminathan (1985) suggests that is is desirable to express weight in relation to some fixed parameter rather than in absolute terms. Weight associated with height (for body length) has been found to be reliable for detecting both under and over nutrition.

To measure the height for adults, a vertical measuring rod or a scale fixed to a wall can be exployed, after removing the shoes, the volunteers were made to stand

on a flat floor by the scale with the feet parallel and with heels, buttocks, shoulders and back of the head touching the upright. The head should be held comfortably erect, with the lower border of the orbit in the same horizontal plane as the external auditory meatus. The arm should be hanging at the sides in a natural manner. The head piece which can be a metal or a wooden block, is gently lowered, crushing the hair, and making contact with the top of the head. These guidelines have been suggested by Jelliffe (1966) for measuring the heights properly.

2. Recording the Health Status through Clinical Examination

According to Roslyn et al (1980), clinical examination and assessment are important adjuncts to dietary and biochemical information as an indication of nutritional status.

Clinical assessment was carried out with the help of a physician and a questionnaire prepared by the investigator. Clinical observation of the tissues of the

eye, skin, hair, face, lips, mucous membranes, tongue, teeth, nails, musculature and thyroid gland were included in the clinical examination. A sample questionnaire used for the purpose is given in Appendix XIV

3. Recording Food and Nutrient Intake

Swaminathan (1985) opines that diet surveys constitute an essential part of any complete study of the nutritional status of individuals or groups, providing essential information on nutrient intake levels, sources of nutrients, food habits and attitudes.

Nutritional well being is a pre-requisite for health, full functional capacity and physical fitness of individuals and populations, according to Shryock(1987).

To determine the food and nutrient intake of the volunteers, dietary and food weighment surveys were conducted, using a 3 day dietary schedule.

The weighment survey was conducted by measuring the raw weights of all the ingredients used for cooking, the total cooked weight and the weight of the cooked food consumed by the individuals. Raw equivalents for the cooked

food was computed and the nutritive value calculated for each individual with the help of food composition tables of ICMR (1984). Those volunteers who complied with the RDA of food and nutrient intake alone were chosen for the study.

The proforma used to record the food intake is given in Appendix X.

4. Recording of Blood Pressure and Pulse Rate

To make sure that the blood pressure and pulse rate of the selected volunteers were normal, these parameters were also recorded.

For measuring the blood pressure, the volunteers were made to lie in a comfortable position and the blood pressure was recorded using a sphymomanometer by auscultatory method (WHO, 1978).

In this method, height of a column of mercury, required to suppress the pulsations in an artery is employed, as a measure of the pressure, in the blood in the artery. To measure the pressure, the cuff was wrapped tightly around the volunteers in the region of the brachial artery. Air is pumped into the cuff

until the air pressure is great enough to compress the artery so that no pulse is heard. The stethoscope is used to listen to the pulse of the brachial artery at the elbow. Then the valve is opened slightly so that the pressure in the cuff begins to fall. Soon a distinct sound is heard as blood spurts into the artery once again. The pressure at this instant is read as the systolic pressure. The sound gets louder and then changes in quality and finally becomes inaudible. Pressure at the time the sound is no longer audible is read as the diastolic pressure.

The pulse was determined by placing the hand lightly on the brachial artery and counting the beats for a minute following the guidelines suggested by Taylor(1980).

IV. Determination of Nutrient Levels in serum

A. Collection of Blood and Separation of serum

10 ml of venous blood was collected from the volunteers using a disposable syringe; 8 ml of the blood was used for separation of serum by centrifugation, and the serum was stored in the refrigerator; 2 ml of the blood was collected in another tube with an anticoagulant added (Potassium

oxalate), for the analysis of haemoglobin, hematocrit and glucose, the above said blood was used.

Serum protein, albumin and globulin, retinol, calcium, zinc and copper and cholesterol were estimated by the investigator in the laboratory within two or three days after the blood samples were collected.

1. Estimation of Serum Iron and Total Iron Binding Capacity

According to Tanphaichitr (1984) for estimating the prevalence of iron deficiency it is preferable to employ a battery of iron parameters in addition to haemoglobin.

Serum iron and total iron binding capacity were estimated using the procedure of Dipyridyl method of Ramsay (1957-1958). Details of the procedure are given in Appendix I.

Unsaturated iron binding capacity was calculated from the formula given below:

UIBC = TIBC - Serum Iron expressed in mcg/dl.

2. Estimation of Total Serum Protein and its Fractions Albumin and Globulin

According to Swaminathan (1985), there is a rapid turnover of plasma proteins and a fall in serum albumin concentration in protein deficiency.

Serum total protein and serum albumin of all the volunteers were determined by using the Biuret method of Hawk and Oser (1965) and quoted by NIN (1983). Details of the procedure are given in Appendix II.

3. Estimation of Serum Retinol

Serum retinol of all the volunteers was estimated using the trifluoroacetic acid method by Neeld & Pearson (1967). The details of the procedure are given in Appendix III.

4. Estimation of Serum Calcium

Serum calcium levels of all the subjects were estimated using the method of Krame and Tisdall given in the National Institute of Nutrition Manual (1983). The details of the procedure are given in Appendix IV.

5. Estimation of Serum Zinc and Copper

The serum zinc and copper values of all the volunteers were determined using the atomic absorption spectrophotometric procedure suggested by Parket et al (1967) and quoted by Varley (1980) using triple acid. The details of the procedure are given in Appendix V.

6. Estimation of Serum Cholesterol

Gertler et al (1984) found no correlation between cholesterol intake and serum cholesterol levels in groups of normal subjects.

Serum cholesterol level was estimated by Zak's method (1954). The Procedure is given in Appendix VI.

B. Analysis of Blood

1. Estimation of Haemoglobin

The haemoglobin levels of all the volunteers was estimated by the Cyanmeth-Haemoglobin method (Dacie and Lewis, 1975) and PCV was determined by the Centrifugation method of NIN (1983) MCHC percent was calculated using the formula given in NIN (1983) manual.

$$\text{MCHC \%} = \frac{\text{Haemoglobin}}{\text{PCV}} \times 100$$

The details of the procedure for the estimation of haemoglobin are given in the Appendix VII and that for PCV percent and MCHC percent in Appendix VIII.

2. Estimation of Blood Glucose

The blood glucose level of all the volunteers was estimated using the Folin-Wu method (1920). The details of the procedure is given in Appendix IX.

V. Analysis of Data

The food and nutrient intake, the anthropometric data, results of clinical examinations and all the values obtained for the biochemical profile were consolidated, analysed and the mean and range were arrived at. An effort was made to compare these values with the values obtained and reported elsewhere in the country and in developed countries. An effort was made to suggest possible guidelines for the biochemical profile of adult men and women.

Results and Discussion

IV. RESULTS AND DISCUSSIONS

The results and discussions pertaining to the present study on the "Biochemical Profile of selected normal healthy adult men and women", are given under the following headings:-

1. Background Details Regarding Volunteers
2. Mean Food and Nutrient Intake of the Volunteers
3. Clinical Examination of volunteers
4. Mean Heights and Weights
5. Biochemical Profile with reference to selected Nutrients.
    a. Mean Haemoglobim, Packed Cell Volume, and Mean Corpuscular Haemoglobin Content Levels.
    b. Mean Serum Iron, total iron binding capacity, unsaturated iron binding capacity and percent transferrin saturation levels.
    c. Mean total serum protein, albumin and globulin levels.
    d. Mean serum retinol levels
    e. Mean serum calcium levels
    f. Mean serum zinc and copper levels
    g. Mean Blood glucose
    h. Mean serum cholesterol levels
6. Mean pulse rate and blood pressure
7. Suggested guidelines for different parameters.

1. Background Details Regarding Volunteers

One hundred and one volunteers, 50 men and 51 women were selected for this study. The volunteers were adults in the age group of 20-39 years. All were educated and belonged to middle or high income group. Their past health condition was good and they were free from hereditary or allergic problems and were not affected by acute or chronic illness. All of them are non-smokers and non-alcoholics.

2. Mean Food and Nutrient Intake of the Volunteers

Table I presents the mean food intake of the volunteers compared against ICMR Recommended Allowances (1984).

TABLE I

MEAN FOOD INTAKE OF MEN AND WOMEN VOLUNTEERS

COMPARED AGAINST ICMR ALLOWANCES (1984)

Food Stuffs	Men			Women		
	Mean Food Intake (g)	ICMR Allow-ances (1984)	Percent Surplus (+) or Deficiency (-)	Mean Food Intake (g)	ICMR Allow-ances (1984)	Percent Surplus (+) or Deficiency (-)
Cereals	412	475	- 13	373	350	+ 7
Pulses	75	65	+ 15	68	70	- 3
Green Leafy Vegetables	58	125	- 58	42	125	- 66
Other Vegetables	75	75	Nil	64	75	- 15
Roots and Tubers	61	100	- 38	62	75	- 17
Fruits	75	30	+150	66	30	+120
Milk and Milk Products	357	200	+ 79	309	200	+ 55
Fats and Oils	40	40	Nil	36	35	+ 3
Sugar or Jaggery	30	40	- 25	30	30	Nil

It is evident from Table I, that a surplus food intake was found with regard to many food stuffs among both men and women volunteers. The mean cereal intake of adult women volunteers was higher by 7 percent than the ICMR allowances (1984). There was a surplus intake of milk and milk products by both men and women, the surplus amounts being 79 percent and 55 percent, respectively, while fat intake was adequate, infact, surplus in the case of women, the intake of sugar was lower by 25 percent in the case of men.

The mean intake of cereals, and roots and tubers was found to be slightly lower than the ICMR Allowances (1984) among the men volunteers, which could be ignored.

The mean intake of roots and tubers and other vegetables were lower among women, when compared with the ICMR Recommended Allowances, which can also be ignored.

However, the intake of green leafy vegetables were definitely lower than the recommended allowances of ICMR (1984). This may be due to the fact that the volunteers did not feel the need for so much leafy vegetables when they were taking good amounts of fruits.

Thus in general the food intakes were quite adequate in both the men and women volunteers.

Details of individual food intake are given in Appendix X and XI for men and women respectively.

Table II shows the mean nutrient intake of the volunteers compared with the ICMR (1984) nutrient allowances for adult men and women.

TABLE II

MEAN NUTRIENT INTAKE OF VOLUNTEERS COMPARED AGAINST ICMR ALLOWANCES (1984)

Nutrients	Men			Women		
	Mean Nutrient Intake (g)	ICMR Allow-ances (1984)	Percent Surplus (+) or Deficit (-)	Mean Nutrient Intake (g)	ICMR Allow-ances (1984)	Percent Surplus (+) or Deficit (-)
Energy (Kcal)	2655	2800	- 5	2538	2200	+ 15
Protein (g)	64	55	+ 16	64	45	+ 43
Calcium (g)	0.85	0.4-0.5	+ 70	0.75	0.4-0.5	+ 50
Iron(mg)	46	20	+ 73	23	30	- 23
Retinol (mcg)	785	750	+ 5	722	750	- 4
Thiamine (mg)	1.5	1.4	+ 7	1.8	1.1	+ 64
Riboflavin (mg)	1.5	1.5	Nil	1.6	1.2	+ 23
Ascorbic Acid(mg)	98	50	+ 97	71	50	+ 44

As presented in Table II, the intake of protein, calcium, thiamine, riboflavin, and ascorbic acid was adequate when compared with the ICMR Allowances (1984) for both men and women. Energy intake was slightly inadequate for men (-5 percent) and retinol intake which was slightly inadequate (-4 percent) for women could be ignored, in view of the fact that these intakes represented intakes only during the selected three days and the intake could have been more on other days as supported through evidences from clinical examination which did not reveal any deficiency symptoms. The iron intake of women was only about three-fourth of the recommended allowances and these women could not be deleted from the study because of the fact that the intake of iron is rather low, in South Indian diets. Thus in general, the food and nutrient intakes were considered adequate, and the volunteers selected for the study.

Individual nutrient intakes are given in Appendix XII for men and XIII for women.

NUTRIENT INTAKE OF MEN AND WOMEN VOLUNTEERS AS PERCENT OF RECOMMENDED ALLOWANCES

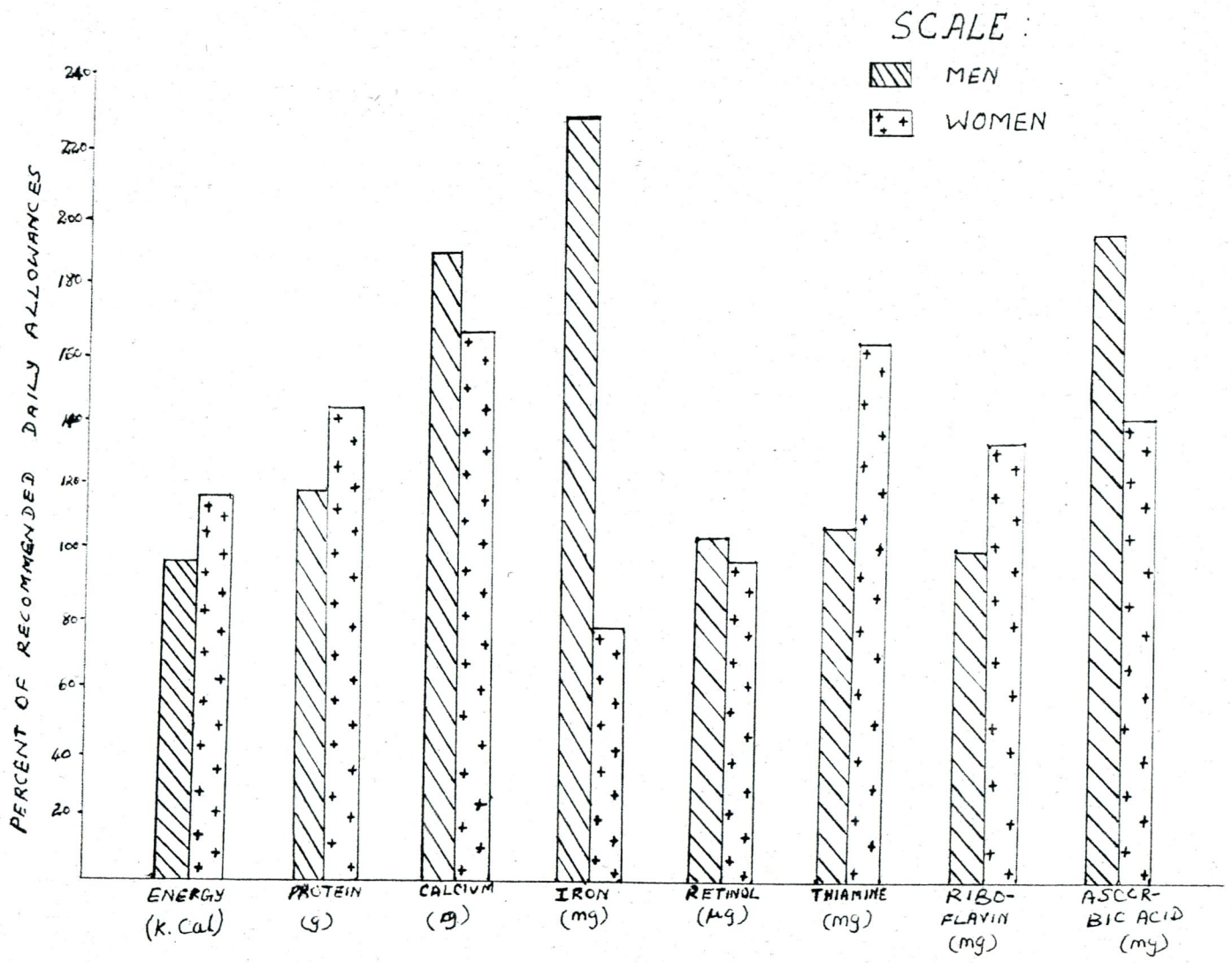

FIGURE : 3

Table III presents the results of the clinical examination carried out on the volunteers.

TABLE III

CLINICAL EXAMINATION OF VOLUNTEERS

S. No.	CRITERIA	Deficiency Symptoms			
		Men		Women	
		N	Percent	N	Percent
1.	HAIR: Easy pluckability, thin and sparse, dull and dry	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
2.	FACE: Moon Face, Paleness, scaling of skin around nostrils	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
3.	EYES: Night blindness, bitots spots, dryness and opacity of cornea, pallor of eye, dry and wrinkled conjuctiva	Nil	Nil	Nil	Nil
	Normal	50	100	51	100

S. No.	CRITERIA	Deficiency Symptoms			
		Men		Women	
		N	Percent	N	Percent
4.	LIPS AND MOUTH: Angular stomatitis, cheilosis, scars and fissures	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
5.	TONGUE: Red, swollen, pale and fissured	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
6.	TEETH: Mottled enamel, caries	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
7.	GUMS: Swollen, spongy, bleeding retracted, receding, loose	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
8.	SKIN: Dryskin, hyperkera-tosis der matitis	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
9.	NAILS: Koilonychia, clubbed	Nil	Nil	Nil	Nil
	Normal	50	100	51	100

S. No.	CRITERIA	Deficiency Symptoms			
		Men		Women	
		N	Percent	N	Percent
10.	GENERAL OEDEMA: Thin and emaciated	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
11.	SOCIAL HABITS: Smoking, Tobacco, chewing, alcoholism	Nil	Nil	Nil	Nil
	Normal	50	100	51	100
12.	Hereditary disorders, ulcer, liver disorder, diabetes-mellitus	Nil	Nil	Nil	Nil
	Normal	50	100	51	100

Clinical examination of the hair, face, eyes, lips, gums, teeth, tongue, skin and nails revealed that none of the volunteers suffered any deficiency symptoms related to the above, and enjoyed good health. It was also clear that they were free from any of the hereditary disorders.

The proforma used for the clinical examination of volunteers is given in Appendix XIV.

Table IV shows the mean heights and weights of the men and women volunteers.

TABLE IV

MEAN AND RANGE OF HEIGHTS AND WEIGHTS OF VOLUNTEERS

Particulars	Men - N - 50		Women - N - 51	
	Height (cm)	Weight (Kg)	Height (cm)	Weight (Kg)
1. Present Study				
Mean	161.7 ± 3.59	60.7 ± 3.7	155.4 ± 4.9	50.8 ± 4.4
Range	155 - 167	54 - 68	144 - 167	45 - 58
2. ICMR (1989)	--	60	--	50

The mean heights of men and women in the present study were 161.7 cm and 155.4 cm respectively. The mean weight recorded for men and women volunteers were 60.7 Kgs and 50.8 Kg. respectively.

The mean values recorded for heights and weights in the present study are in tune with the values suggested for the reference man and woman, and hence the selected men and women represent the normal group on whom biochemical profile could be done.

The individual values of heights and weights for adult men and adult women are given in Appendix XV.

5. Biochemical Profile with reference to selected Nutrients

a. Mean Haemoglobin, Packed Cell Volume and Mean Corpuscular Haemoglobin content levels

Table V shows the mean and range of haemoglobin, Packed Cell Volume and mean corpuscular haemoglobin content among the volunteers.

TABLE V

MEAN AND RANGE OF THE HAEMOGLOBIN, PACKED CELL VOLUME AND MEAN CORPUSCULAR HAEMOGLOBIN CONTENT OF VOLUNTEERS

Particulars	Haemoglobin		PCV Percent		MCHC Percent	
	Men A	Women B	Men A	Women B	Men A	Women B
1. Present study						
Mean	14.8 ± 0.74	12.2 ± 1.4	40.5 ± 1.9	34.7 ± 2.3	36.5 ± 1.8	35.1 ± 3.7
Range	13.1 -16.3	10.2 -14.6	34 -48	32 -39	33.8 -40.4	30.3 -44.1
't' Value A Vs. B	9.2*		6.5*		2.4**	
2. Swaminathan (1985)	14-16	13-15	44.0-48.0	40.0-44.0	34	36
3. Tietz(1987)	13.5-17.5	12.0-16.0	Not Available			
4. NIN (1978)	12.79	11.64	Not Available			

* Significant at 1% level

** Significant at 5% level

The mean haemoglobin levels obtained for men and women were found to be 14.8g/dl and 12.2g/dl, respectively. The haemoglobin levels for adult women were comparable with the values given by the National Institute of Nutrition (1978) and with these given by Swaminathan (1974) and Tietz (1987).

The mean PCV percent obtained for men and women were 40.5 and 34.7 percent respectively. These values were slightly lower than the values given by Swaminathan (1974), i.e. 44.0 - 48.0 for adult men and 40.0 - 44.0 for adult women.

For men and women, the MCHC percent obtained were 36.5 and 35.1 percent respectively. These values were comparable with values suggested by Swaminathan (1974).

The difference in haemoglobin levels and PCV between men and women were found to be statistically significant at 1% level, while the difference in MCHC percent between men and women was significant at 5% level.

The individual values of haemoglobin, PCV percent and MCHC of volunteers are given in Appendix XVI.

Table VI highlights the mean and range values for serum iron, total iron binding capacity, unsaturated iron binding capacity and percent saturation of transferrin for men and women volunteers.

TABLE VI

MEAN AND RANGE OF SERUM IRON, TIBC, UIBC AND PERCENT SATURATION OF TRANSFERRIN OF VOLUNTEERS

Particulars	Serum Iron mcg/dl		TIBC mcg/dl		UIBC mcg/dl		Percent Transferrin saturation	
	Men A	Women B	Men A	Women B	Men A	Women B	Men A	Women B
1. Present Study								
Mean	117 ±30.9	107.8 ± 23.6	293 ± 49.1	294 ± 37.8	176 ± 27.1	186 ± 29.9	39.5 ± 5.9	36.5 ± 6.4
Range	44-165	56.6-147	153-350	153-348	96.4-218.8	96.5-235	30.6-49.3	22.2-62.5
't' value A Vs B	1.34^{NS}		0.12^{NS}		1.76^{NS}		2.4**	
2. Swaminathan (1974)	50-180		300	450	Not Available		Not Available	
3. Varley (1980)	80-175	60-160	253-416	250-416	Not Available		33	47
4. Tietz (1987)	65-170	50-170	250	450	Not Available		20-55	

** - Significant at 5% level

NS - Not significant.

The mean serum iron levels obtained for men and women were 117 mcg/dl and 107.8 mcg/dl respectively. The range of values for men were 44-165 mcg/dl, for women, 56.6 - 147 mcg/dl; while these values were comparable with the values quoted by Varley (1980) and Tietz (1987), the ranges offered by these scientists are much wider than the ranges suggested in the present study.

For TIBC, the mean values for men and women were 293 and 294 mcg/dl. Tietz (1987) has given a wider range for TIBC, as 250-450 mcg/dl than the range suggested in the present study.

For percent saturation of transferrin, Tietz (1987), suggested a wide range of 20-55 mcg/dl for both men and women against 30.6-49.3 and 22.2-62.5 mcg/dl suggested for men and women respectively in the present study.

The difference in percent transferrin saturation between men and women were found to be statistically significant at 5% level.

Individual values of serum Iron, TIBC, UIBC and percent transferrin saturation are given in Appendix XVII.

MEAN OF SERUM IRON AND PERCENT SATURATION OF MEN AND WOMEN VOLUNTEERS

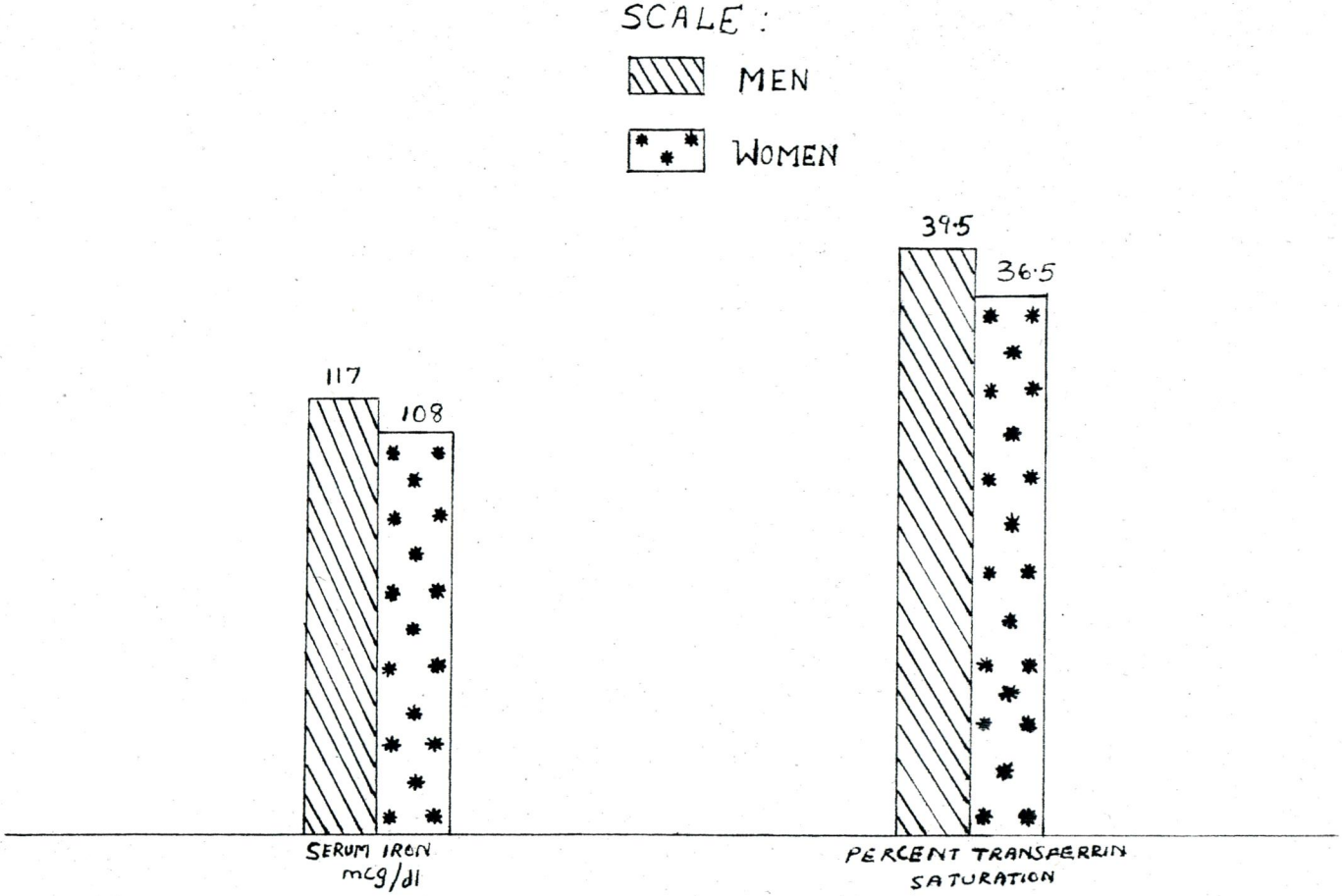

FIGURE - 4

Table VII shows the mean and range levels of total protein, albumin and globulin of the volunteers.

TABLE VII

MEAN AND RANGE OF TOTAL PROTEIN, ALBUMIN AND GLOBULIN OF VOLUNTEERS

Particulars	Total Protein		Albumin		Globulin	
	Men A	Women B	Men A	Women B	Men A	Women B
1. Present Study						
Mean	6.95 ±0.57	6.78 ±0.54	5.1 ±1.23	4.6 ±0.56	2.5 ±0.42	2.2 ±0.50
Range	5.9-7.8	5.8-8.0	3.5-5.8	3.5-6.1	1.5-3.5	1.3-3.6
't' value A Vs B	1.55^{NS}		2.63*		3.3*	
2. Swaminathan (1974)	6-8		4-5.5		Not given	
3. Varley (1980)	5.8-7.8		3.5-5.6		2.6-3.1	
4. Tietz (1987)	6.4-8.3		3.5-5.0		Not given	

* Significant at 1% level

NS Not Significant.

It is evident from Table VII that the mean total protein values were 6.95g/dl for men and 6.78g/dl for women. The range levels for men and women were 5.9-7.8g/dl and 5.8-8.0g/dl, respectively. The range values obtained in the present study is comparable with the range values reported by Varley (1980), Tietz (1987) and Swaminathan (1974).

The mean albumin for men and women in the present study were 5.1g/dl and 4.6g/dl respectively. The range values were 3.5-5.8g/dl for men and 3.5-6.1g/dl for women. These values were comparable with the values suggested by Varley (1980).

The mean globulin levels were 2.5g/dl and 2.2g/dl for men and women respectively. The range values obtained were 1.5-3.5g/dl for men and 1.3-3.6 for women. The range values obtained for globulins in the present study appear to be slightly higher than the range values indicated by Varley(1980).

The difference in albumin and globulin levels between men and women were found to be statistically significant at 1% level.

Individual values for total serum protein, albumin and globulin levels are given in Appendix XVIII.

MEAN OF TOTAL SERUM PROTEIN, ALBUMIN AND GLOBULIN OF MEN AND WOMEN VOLUNTEERS

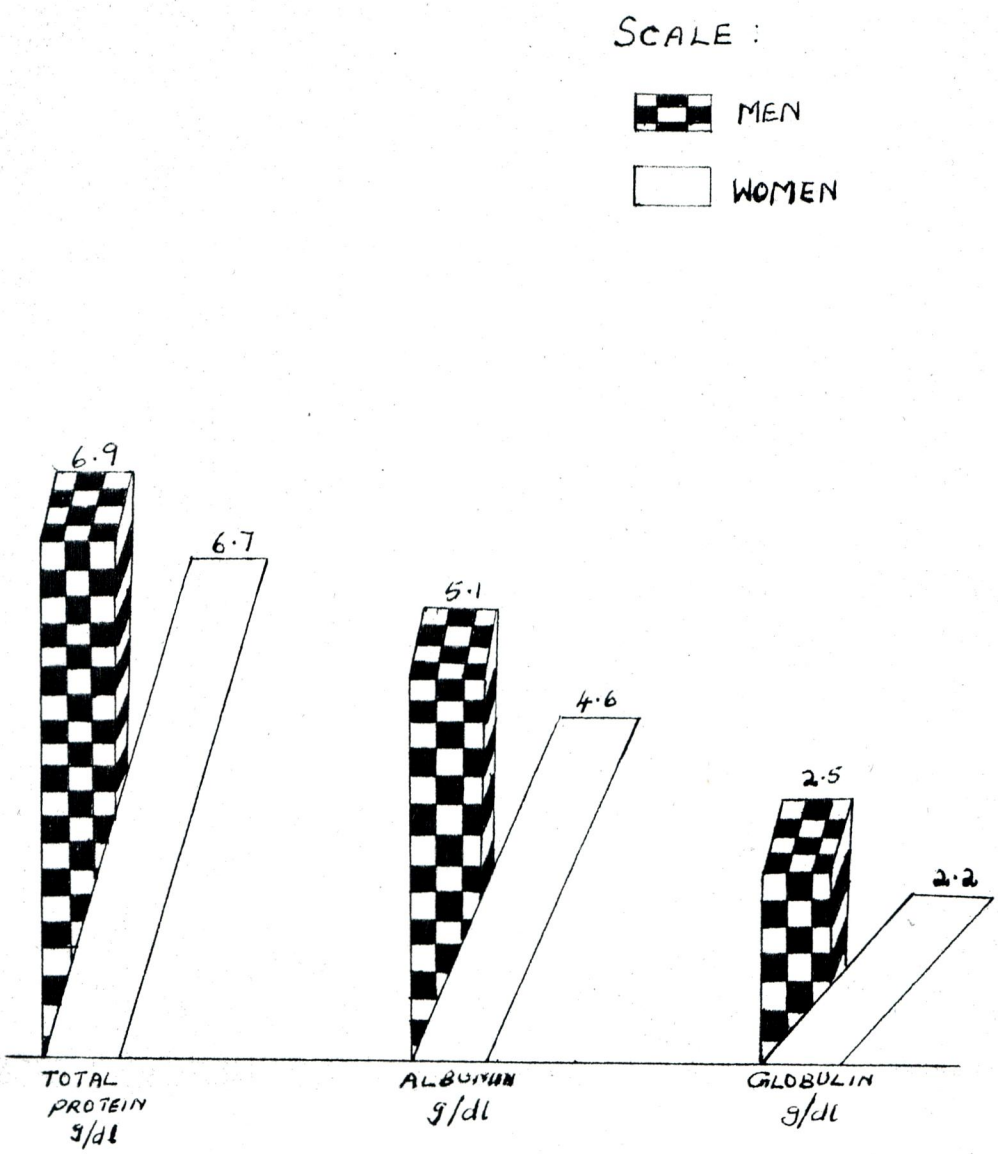

FIGURE - 5

Table VIII shows the mean, range levels of serum retinol among men and women volunteers of the present study.

TABLE VIII

MEAN AND RANGE VALUES OF SERUM RETINOL OF VOLUNTEERS

Particulars	Serum Retinol mcg/dl		
	Men A		Women B
1. Present study			
Mean	40.58±6.99		38.28±5.8
Range	28.7 - 54.5		28.6 - 53.7
't' value A Vs B		1.79^{NS}	
2. Swaminathan (1974)		25-90	
3. Varley (1980)		20-50	
4. Tietz (1987)		30-65	

NS Not significant.

From Table VIII, it may be noticed that the mean retinol levels for men and women were 40.58 mcg/dl and 38.28 mcg/dl, respectively. The range values obtained were 28.7-54.5 mcg/dl for men and 28.6-53.7 mcg/dl for women. Tietz (1987) suggested a value of 30-65 mcg/dl which is in tune with the findings of the present study. The values of the present study were also in tune with the values obtained by Swaminathan (1974) and Varley (1980).

The values recorded by women were lower than the values recorded by men. Individual values of serum retinol are given in Appendix XIX.

Table IX gives the mean and range levels of serum calcium among the volunteers.

TABLE IX

MEAN AND RANGE VALUES OF CALCIUM OF VOLUNTEERS

Particulars	Serum Calcium mg /dl	
	Men A	Women B
1. Present study		
Mean	10.36±0.64	10.69±0.62
Range	8.9 - 11.6	9.5 - 11.9
't' value A vs B	2.6*	
2. Swaminathan (1974)	9-11	
3. Varley (1980)	9-11	
4. Tietz (1987)	8.4-10.2	

* Significant at 1% level.

It is evident from Table IX that the mean calcium levels for men and women were 10.36 mg/dl and 10.69 mg/dl, respectively. The range values were 8.9-11.6 mg/dl for men and 9.5-11.9 mg/dl for women. The mean and range levels of serum calcium for adult men and women were comparable with the values quoted by Swaminathan (1974), Varley (1980) and Tietz (1987).

In this particular parameter, the upper limit of range values for men and women were found to be slightly higher when compared with the values given by Varley (1980) and Tietz (1987).

The difference in serum calcium levels between men and women were found to be statistically significant at 1% level. Individual values of serum calcium are given in Appendix XIX.

Table X presents the mean and range levels for serum zinc and Copper of Volunteers.

RANGE OF SERUM CALCIUM OF MEN AND WOMEN VOLUNTEERS

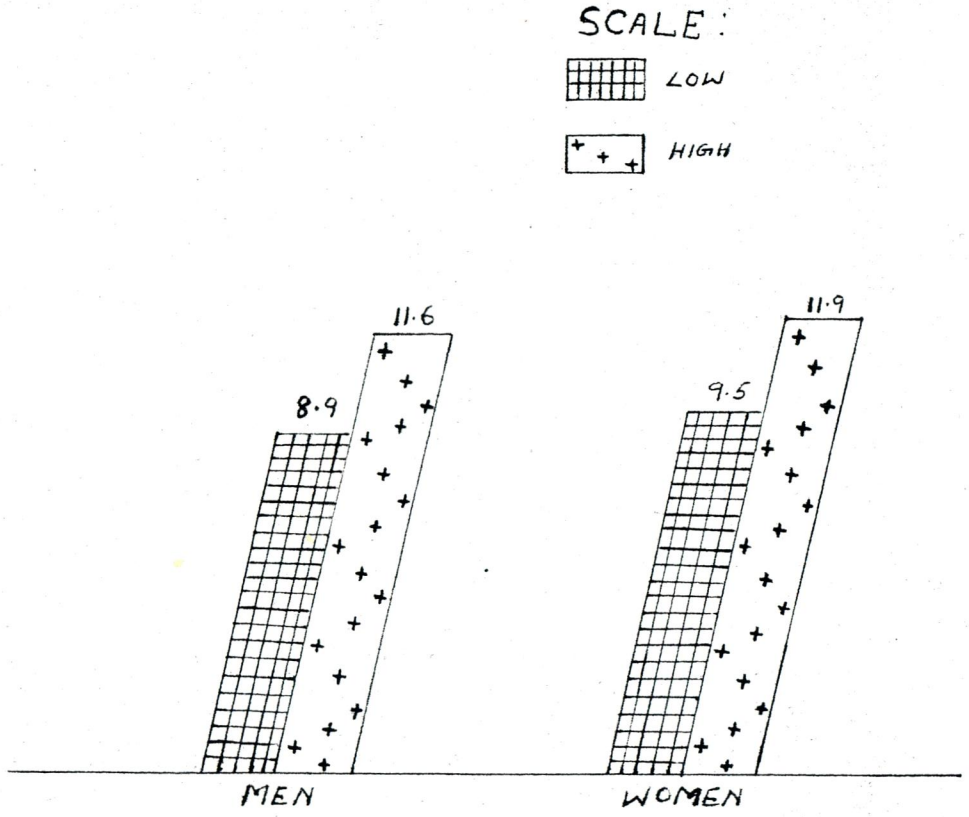

FIGURE-6

TABLE X

MEAN AND RANGE LEVELS OF ZINC AND COPPER OF THE VOLUNTEERS

Particulars	Serum Zinc mcg/dl		Serum Copper mcg/dl	
	Men A	Women B	Men A	Women B
1. Present Study				
Mean	95.7±11.5	93.12±14.95	104±13.9	99.7±16.4
Range	67-119	60-120	85-137	72-131
't' value A vs B	0.96^{NS}		1.42^{NS}	
2. Swaminathan (1974)	120-130		100	
3. Varley (1980)	72-115		75-160	
4. Tietz (1987)	70-150		70-140	80-155

NS = Not significant.

The mean and range levels of serum copper obtained for men were 104 mcg/dl and 85-137 mcg/dl, respectively. The mean and range values of serum copper among adult women were 99.7 mcg/dl and 72-131 mcg/dl respectively. The range of serum copper levels were compared against the

values reported by Swaminathan (1974), Varley (1980) and Tietz (1987). As is evident from Table X the ranges suggested by Varley (1980) and Tietz (1987) were more flexible than the range suggested in the present study and the values given by Swaminathan (1974).

Table X shows the mean and range level of serum zinc as 95.7 mcg/dl for men and 93.1 mcg/dl for women. The range of serum zinc levels were 67-119 mcg/dl and 60-126 mcg/dl for men and women, respectively. These values were comparable with the levels quoted by Swaminathan (1974) and Tietz (1987). However, the lower limits were found to be slightly lower and the higher limits were slightly higher than the values reported in the present study.

Individual values of serum zinc and serum copper are given in Appendix XX.

Table XI shows the mean and range values of blood glucose of the volunteers.

TABLE XI

MEAN AND RANGE VALUES OF BLOOD GLUCOSE OF THE VOLUNTEERS

Particulars	Blood Glucose mg/dl	
	Men A	Women B
1. Present Study		
Mean	98.5 ± 11.7	95.01 ± 11.7
Range	76 - 120	74 - 120
't' value A vs B	1.49^{NS}	
2. Swaminathan (1974)	80-120	
3. Varley (1980)	70-120	
4. Tietz (1987)	120	

NS - Not significant.

It is evident from Table XI, that the mean and range of blood glucose values of adult men were 98.5 mg/dl and 76-120 mg/dl, respectively. For adult women, the mean and range of blood glucose levels were 95.01 mg/dl and 74-120 mg/dl, respectively. These values were comparable with values reported by Swaminathan (1974), Varley (1980) and Tietz (1974).

The difference in the values registered for blood glucose was not significant statistically. Individual values of blood glucose are given in Appendix XXI.

Table XII shows the mean and range levels of serum cholesterol of the volunteers.

TABLE XII

MEAN AND RANGE VALUES OF SERUM CHOLESTEROL OF VOLUNTEERS

Particulars	Serum Cholesterol mg/dl	
	Men A	Women B
1. Present study		
Mean	176.7 ± 28.68	170.6 ± 34.8
Range	125 - 238	104 - 240
't' value A Vs B	0.96^{NS}	
2. Swaminathan (1974)	120-260	
3. Varley (1980)	150-240	
4. Tietz (1987)	140-220	

NS - Not significant.

Table XII reveals the mean serum cholesterol levels for men and women as, 176.7 mg/dl and 170.6 mg/dl, respectively. The range levels of cholesterol are 125-238 mg/dl for men and 104-240 mg/dl for women. These values were comparable with the values reported by Varley (1980), Swaminathan (1974) and Tietz (1987) except the fact that the ranges suggested by Varley (1980) and Tietz (1987) were rather wide.

The difference in serum cholesterol levels between men and women were not significant statistically.

Individual values of serum cholesterol levels are given in Appendix XXI.

Table XIII shows the mean pulse rate and blood pressure of the volunteers.

TABLE XIII

MEAN PULSE RATE AND BLOOD PRESSURE OF THE VOLUNTEERS

Particulars	Pulse Rate Per minute		Blood Pressure mm/Hg	
	Men A	Women B	Men A	Women B
1. Present study				
Mean	72	72	120/80	118/80
2. Best & Taylor (1980)	72	72	120/80	120/80

The mean pulse rate of the volunteers was 72/minute for both men and women volunteers. The blood pressure recorded was 120/80 mm/Hg for men and 118/80 for women. These values were comparable with the values given by Best and Taylor (1980).

Table XIV gives the desirable mean levels of different parameters for normal healthy adult men and women.

TABLE XIV

SUGGESTED MEAN VALUES FOR DIFFERENT PARAMETERS

	Nutrients	Suggested Mean			
		Present Study		Varley (1980)	
		Men	Women	Men	Women
1.	Haemoglobin (g/dl)	14.8	12.2	-	-
2.	Packed Cell Volume (Percent)	41	35	-	-
3.	Mean Corpuscular Haemoglobin content (Percent)	37	35	-	-
4.	Serum Iron (mcg/dl)	117	108	80-175	60-160
5.	Total Iron Binding Capacity (mcg/dl)	293	294	253-416	250-416
6.	Unsaturated Iron Binding capacity (mcg/dl)	176	186	-	-
7.	Transferrin saturation(Percent)	40	37	33	47
8.	Total Serum Protein (g/dl)	6.95	6.78	5.8 -	7.8
9.	Serum Albumin(g/dl)	5.1	4.6	3.5 -	5.6
10.	Serum Globulin (g/dl)	2.5	2.2	2.6 -	3.1
11.	Serum Retinol (mcg/dl)	40.6	38.3	20 -	50
12.	Serum calcium (mg/dl)	10.4	10.7	9 -	11
13.	Serum Zinc (mcg/dl)	95.7	93.1	72 -	115
14.	Serum Copper (mcg/dl)	104	99.7	75 -	160
15.	Blood Glucose(mg/dl)	98	95	70 -	120
16.	Serum Cholesterol (mg/dl)	176	170	150 -	240

As evident in Table XIV, the mean values of serum Iron, TIBC, total serum protein, albumin, retinol, calcium, zinc and copper, blood glucose and serum cholesterol of the volunteers are comparable with the range quoted by Varley (1980), for the respective parameters. However, the mean level of transferrin saturation in the present study for men were higher and for women lower than the values given by Varley (1980) for the same. Serum Globulin levels were found to be lower in the present study when compared with the values given by Varley (1980).

Table XV suggests the range levels of different parameters for health adult men and women, which may be used for comparison purposes.

Summary and Conclusion

V. SUMMARY AND CONCLUSION

The present study was undertaken with the aim of identifying healthy adult men and women and finding their biochemical profile with regard to selected nutrients, so as to arrive at guidelines for biochemical profile of normal adult men and women. The present study included the nutritional status of 50 men and 51 women volunteers doing moderate activity and in the age group of 20 - 39 years.

Background details regarding the volunteers were collected. Clinical examination was carried out for all the volunteers to ensure that they were not suffering from any deficiency diseases. The food and nutrient intake of the volunteers was assessed using food weighment method and comparing the intakes of volunteers with the food and nutrient allowances recommended by the Indian Council of Medical Research (ICMR, 1984).

Based on the results obtained by conducting the clinical examination and food weighment surveys, 10 ml of blood was drawn from the volunteers and used for the estimation of haemoglobin, PCV, MCHC and glucose using

whole blood, serum iron, TIBC, UIBC, percent saturation of transferrin, total protein, albumin and globulin, retinol, calcium, zinc and copper, cholesterol, after separating the serum. The results obtained from the above were compared with the available guidelines of Swaminathan (1974), Varley (1980) and Tietz (1987).

The mean and range values obtained in the present study for men and women volunteers are presented as follows:

Haemoglobin values for men ranged from 13.1 - 16.3g/dl and for women were 10.2 - 14.6g/dl, while the means were found to be 14.8g/dl for men and 12.2g/dl for women. The values of mean and range for PCV percent and MCHC percent for men were 40.5 and 34-48 percent and 36.5 and 33.8-40.4 percent respectively. The PCV and MCHC values for women were 34.7 and 32-39 percent and 35.1 and 30.3 - 44.1 percent respectively.

The mean values for serum iron, TIBC, UIBC and percent saturation of transferrin values obtained for men were

117 mcg/dl, 293 mcg/dl, 176 mcg/dl and 39.5 percent, respectively and corresponding values for women were 107.8 mcg/dl, 294 mcg/dl, 186 mcg/dl and 36.5 percent, respectively.

The range levels for serum iron, TIBC, UIBC and percent saturation of transferrin for men were found to be 44-165 mcg/dl, 153-350 mcg/dl, 96 -218 mcg/dl and 30.6 - 49.3 percent, respectively and corresponding values for women were 56.6 - 147 mcg/dl, 153 - 348 mcg/dl, 96.5 - 235 mcg/dl and 22.2 - 62.5 percent, respectively.

The serum protein, albumin and globulin levels of men were analysed and the range found as 5.9 - 7.8g/dl, 3.5 -5.8g/dl and 1.5-3.5g/dl with respective means of 6.95g/dl 5.1g/dl and 2.5g/dl while those for women were 5.8 - 8.0g/dl 3.5 - 6.1g/dl and 1.3-3.6g/dl with means of 6.78g/dl, 4.6g/dl and 2.2g/dl, respectively.

The ranges for serum calcium levels for both men and women in the present study were 8.9 - 11.6mg/dl and 9.5-11.9mg/dl, respectively. Women recorded a higher mean value of 10.69 mg/dl in comparison to 10.36mg/dl recorded by men.

Serum Copper levels ranged from 85 - 137 mcg/dl for men and 72-131 mcg/dl for women, and their mean values were 104mcg/dl and 99.7 mcg/dl for men and women, correspondingly. For serum zinc, the range values were 67-119 mcg/dl for men and 60-126 mcg/dl for women and their mean values were 95.7 mcg/dl for men and 93.12mcg/dl for women.

The range levels of blood glucose for men were 76-120 mg/dl for men and 74-120 mg/dl for women. The corresponding mean values were 98.5mg/dl and 95.01mg/dl and women.

The findings of the present study with reference to serum cholesterol were 125-238mg/dl for men and 104-240mg/dl for women. The mean values were 176.8mg/dl for men and 170.6mg/dl for women.

Results obtained from this study lead to the following conclusions:

1. Biochemical profile of selected, healthy, adult men and women are comparable with the available western standards with regard to albumin, serum zinc and copper, serum cholesterol, blood glucose and calcium whereas MCHC percent and serum calcium are found to be higher. However, PCV percent, haemoglobin and globulin were found to be slightly lower than the available western standards.

2. Serum calcium and TIBC are found to be higher in women than men.

Based on the above conclusions, it is recommended, that it is imperative to develop local guidelines on biochemical profile of normal healthy adult men and women. Further studies along these lines may help in developing such guidelines.

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Appendix

APPENDIX I

ESTIMATION OF SERUM IRON, TIBC, UIBC AND TRANSFERRIN SATURATION PERCENT: RAMSAY'S DIPIRYDIAL METHOD

1. DIPIRYDYL METHOD: (Ramsay, 1954, 1958).

Ferrous iron gives a pink colour with dipyridyl. A solution of dipyridyl in acetic acid is added to serum followed by a reducing agent, proteins are removed by heating in boiling water and then centrifuging or filtering.

REAGENTS

1. Dipyridyl, 0.1% in acetic acid 3 percent v/v
2. Sodium sulphite, 0.1m. Dissolve 1.26 gms of anhydrous sulphite or 2.52 gms of Na_2SO_3. 7 H_2O in water and make up to 100ml prepare freshly every few days.
3. Chloroform
4. Standard solution containing 100 gms iron per ml. Dissolve 0.498 grms of ferrous sulphate in water add 1 ml.
   Concentrated H_2 SO_4 and make to a litre.
   Alternatively use a solution of ferrous ammonium sulphate $(NH_4)_2$ SO_4, $FeSO_4$, $6H_2O$ Containing 70.2 mg/litre.

TECHNIQUE

Mix equal volumes of serum, p.1m sodium sulphate and dipyridyl reagent in a glass stoppered tube which can be centrifuged. Heat in boiling water for five minutes. Cool, add 1 ml of chloroform, stopper and shake vigorously for thirty seconds. Remove the stopper and centrifuge for five minutes at 30 r.p.m. If the supernatent fluid is not completely clear repeat the shaking and centrifuging. Read at 520 millimicrons or using a green filter. As blank use water instead of serum. For the standard put through the working standard in the same way.

Clean tubes used by placing there in boiling 5 MHCL. Then wash with distilled water and keep for this determination duly.

CALCULATION

Microgram iron per 100ml of serum =

$$\frac{\text{Reading of unknown} \times 300}{\text{Reading of standard}}$$

The readings are linear with concentration to atleast 500mg per 100ml. To obtain a calibration curve dilute 5 ml of the stock standard to 100ml with water and

set up tubes containing 0.4, 0.8, 1.2, 1.6 and 2.0ml of this make each to 2ml with water and develop the colour as described above and red against the blank. These correspond to 100, 200, 300, 400 and 500mg/100ml.

If the iron binding capacity is being done by Ramsay's method at the same time as serum iron it may be more convenient to use the double strength sulphite and dipyridyl reagent for both. Then for serum iron use 1 volume of serum, 1 volume of water and 0.5 volume of each of 0.2m sulphate and 0.2% dipyridyl in 3 percent acetic acid.

TIBC RAMSAY'S DIPYRIDYL METHOD

REAGENTS

1. Ferric chloride solution 5 mg iron per ml in 0.005 NHCL. Prepare a stock solution containing 145mg of $FeCl_3$ per 100ml of 0.5N acid and dilute 1 to 100 with water.
2. Magnesium Carbonate "light" for absorption
3. Sodium sulphite (0.2m) 2.52grams of the anhydrous salt per 100ml.
4. Dipyridyl (0.2 percent) in acetic acid 3 percent v/v chloroform and standard solutions as for the method of serum iron.

TECHNIQUES

Add 4 ml of the ferric chloride solution to 2ml of serum. After standing for five minutes add 400mg of Magnesium carbonate (100mg for each ml of ferric chloride) shake frequently and vigorously for thirty to sixty minutes. Centrifuge and pipette of 4ml of the supernatent fluid for iron determination. If the dipyridyl method is used to add 1 ml each of the 0.2m sulphite and 0.2 percent dipyridyl and proceed as described previously for determining serum iron. The result gives the total iron binding capacity. If the serum iron is determined at the same time the percentage saturation is easily calculated.

CALCULATION

Since in this case the volume of serum in the 4ml of supernatent fluid is 1.33ml if the same proportions are used for the standard as in Note (i.e. 2 ml of the standard containing 3 mg per iron per ml. 2 ml water and 1 ml each of dipyridyl and sulphite).

TIBC in microgram/.00ml solution.

$$= \frac{\text{Reading of unknown x 540}}{\text{Reading of standard}}$$

UNSATURATED IRON BINDING CAPACITY(UIBC)

= TIBC - Serum Iron (Mcg/dl)

PERCENT SATURATION OF TRANSFERRIN

$$= \frac{\text{Serum Iron}}{\text{TIBC}} \times 100$$

APPENDIX II

ESTIMATION OF TOTAL SERUM PROTEIN, ALBUMIN AND GLOBULIN

BIURET METHOD

AIM

To estimate the total protein, albumin and globulin in serum.

PRINCIPLE

Proteins form a purple coloured complex with cerpric irons in alkaline solution. The reaction takes its name from the simple compounds, which reacts in the same way.

REAGENTS

1. Sodium Chloride dilutent:

Dissolved 9gms of sodium chloride in water and made upto 1 litre.

2. Stock Bierect reagent

Dissolved 45gms of potassium sodium tartarate (Rochelle's Salt) in 400ml of 0.2N sodium hydroxide. Added 15gms of cupric sulphate stirred until it dissolved. Added 5gms of potassium iodide and diluted to one litre with 0.2N Sodium hydroxide.

3. Dilute Biuret Reagent

Diluted 200ml of stock biuret reagent to 1 litre with 0.2N sodium hydroxide and added 5gms of potassium iodide.

4. Standard albumin solution:

Weighed 400mg albumin and dissolved in 0.8 percent saline solution. Made up the volume 100ml with sodium hydroxide so that 1.0ml of this solution contains 4mg of proteins.

5. 22.5 percent sodium sulphite solution.

PROCEDURE

Into a series of test tubes 0.5 - 2.5ml of standard albumin solution was pipetted out and then the volume made upto 3 ml with water. Into another test tube pipetted out 0.2ml of serum and dilute with 0.9 percent saline upto 5ml. From this 2ml of the solution was taken, made upto 3 ml with water and treated as unknown. Now added 3ml of dilute biuret reagent to all test tubes. Along with this a blank was prepared. The colour developed was read colorimetrically at 500gm after 30 minutes. The amount of protein in the serum was calculated. This gives the total volume.

PRECIPITATION OF GLOBULINS

Globulin is precipitated by mixing 0.2ml of serum with 4.8ml of 22.5 percent sodium sulphite solution. Stoppered the tubes are inverted it several times and left in the incubator at 40°C overnight. Filtered the solution next day using whatman No.1 filter paper.

Taken 2 ml of the filtrate and carried out the experiment as for total proteins. The concentration in gm per albumin present is determined from the standard graph.

Globulin value can be obtained by deducting albumin volume from the total proteins.

APPENDIX III

ESTIMATION OF SERUM VITAMIN A (RETINOL)

The serum Vitamin A was estimated by the trifluoro acetic acid method by Neeld and Pearson as modified and suggested by Roch et al, quoted by Gyorgy and Pearson.

1. Absolute ethanol: Purified for spectrophotometry.
2. n-Hexane: Fischer certified reagent special for spectrophotometry.
3. Chloroform: Merck neagent special for spectrophotometry
4. Trifluroroacetic acid: Reagent grade (Sigma)
5. In alcoholic potassium hydroxide
6. Stock Vitamin-A: 344 mcg of Vitamin-A acetate (300 mg of Vitamin-A) was dissolved in chloroform and made upto 100ml, 1.0ml of the stock contains 3000 mcg of retinol.
7. Working standard

a. 0.1 ml of stock standard diluted to 10ml with chloroform (30 mcg/ml).

b. 0.1 ml of stock diluted to 5 ml with chloroform (60 mcg/ml).

c. 0.15 ml of stock diluted to 5 ml with chloroform (90 mcg/ml).

d. 0.1 ml of stock diluted to 2.5 ml with chloroform (120 mcg/ml).

Each intermediate standard was again diluted in the ratio 1:10 and from each standard finally 1 ml was taken.

TECHNIQUE

The serum extract (0.5ml or less) was saponified with an equal volume of IN enthanalic potassium hydroxide in a water bath at 60°C for 20 minutes. The mixture was called and vigorously shaken in a glass stoppered tube with an equal volume (1ml) of n-Hexane for 10 minutes. The tube was centrifuged for 1 minute at 100g to separate the layers. An aliquot (0.8ml) was pipetted off for the determination of retinol. The n.Hexane was evaporated from this aliquot in a water-bath at 60°C in a stream of oxygen free nitrogen. The last traces of n-hexane were removed by nitrogen blowing at room temperature. The residue was taken up in (0.5ml) chloroform; 1 drop of acetic anhydride was added followed by 0.1ml of trigluoroacetic acid. The mixture is shaken vigorously and the optical density at 620 mu was determined exactly 30 seconds after addition of the trifluoroacetic acid.

APPENDIX IV

ESTIMATION OF SERUM CALCIUM

PRINCIPLE

Calcium is precipitated as oxalate and is titrated with standard potassium permanganate.

REAGENTS

1. 4% ammonium oxalate solution
2. Dilute ammonia solution (2ml of liquor ammonia + 98ml of water)
3. 1 N Sulphuric acid
4. 0.01N potassium permanganate solution
5. 0.01N oxalic acid: sodium oxalate is dried in an oven at 100-150^{o}C for 12 hour. Exactly 0.67g is dissolved in redistilled water 5 ml. Conc H_2SO_4 is added and solution made upto 1 lit. after it has called down.

Standardisation of potassium permanganate solution 25ml of 0.01N oxalic acid is transferred to an Erlenmeyer flask. One ml of concentrated H_2 SO_4 is added, warmed to about 70^{o}C and titrated against $KMno_4$ solution, till the faint pink colour remains.

PROCEDURE

2ml of the sample is taken into a 15ml centrifuge tube. Add 2ml of distilled water and 1 ml of 4% ammonium oxalate solution and mix thoroughly and leave overmight. Again the contents one mixed and centrifuged for 5 mins at 1500 rpm. The supernatent liquid is poured off and the centrifuge tube drained by inverting the tube for 5 min on a rack (care should be taken not to disturb the precipitate). The mouth of the centrifuge tube is wiped with a piece of filter paper. The precipitate is stirred and the sides of the tubes are washed once more with 3ml dilute ammonia. It is centrifuged again and drained as before. The precipitate is washed once more with dilute ammonia to ensure the complete removal of ammonium oxalate. The precipitate is dissolved in 2ml of IN H_2SO_4. The tube is heated by placing it in a boiling water bath for 1 min and titrated against 0.01N $KMnO_4$ solution to a definite pink colour persisting for atleast 1 min.

CALCULATION

1 ml of 0.01N $KMnO_4$ is equivalent to 0.2004 mg of calcium.

mg of calcium/100ml serum = (x-6) x 0.2004 x 100/2 where x = no. of ml of 0.01N $KMnO_4$ required to titrate 2 ml of H_2SO_4 (Blank).

If the normality of $KMnO_4$ is N, the value obtained in the above formula should be multiplied by the factor N/0.01.

NOTE: $KMnO_4$ soln. needs to be frequently standardized.

APPENDIX V

ESTIMATION OF SERUM ZINC AND COPPER

One ml of the sample to be analysed was digested with 10ml of triple acid (nitric, sulphuric, perchloric acids in the ratio 9:2:1) and made upto 50ml. The made up sample was then fed into the atomic absorption spectrophotometer for the analysis of Cu and Zn.

APPENDIX VI

ESTIMATION OF SERUM CHOLESTEROL - ZAK'S METHOD

PRINCIPLE

Cholesterol reacts with ferric chloride in the presence of concentrated sulphuric acid to give a pink colour. The intensity of the colour developed is directly proportional to the amount of cholesterol present and is read at 540mu in a colorimeter.

REAGENTS

1. Stock Ferric chloride solution:

840 mgs of pure ferric chloride was weighed and dissolved in glacial acetic acid and made upto 100ml with the same.

2. Ferric chloride precipitating reagent

10ml of the stock ferric chloride reagent was placed in a 100ml standard flask and made upto the mark with pure acetic acid.

3. Ferric chloride diluting reagent

8.5 ml of stock solution of ferric chloride was diluted to 100ml with pute glacial acetic acid in a 100ml standard flask.

4. Standard cholesterol solution:

100mg of pure cholesterol standard was placed in 100 ml standard flask containing 0.85 ml of ferric chloride stock reagent; made upto the mark with glacial acetic.

5. Working standard:

10ml of stock standard was made upto 100ml with acetic acid. 1ml of solution had 100 of cholesterol.

PROCEDURE

0.5-2.5 ml of the working cholesterol solution was pipetted out into clean dry test tubes. 1 ml of this solution contains 100 of cholesterol. The total volume of each tube was made upto 5 ml with ferric chloride diluting reagent.

To 0.1ml of the serum added 4.9 ml of ferric chloride precipitating reagent and mixed well. Allowed to stand for a while and centrifuged. Transferred 2.5 ml of the clear supernatent in a dry test-tube and added 2.5ml of ferric chloride diluting reagent. Mixed well. The tubes were kept in cold water. To each tube added 4ml of concentrated sulphuric acid drop by drop. The solution was mixed well. The tubes wereaallowed to come to room temperature. A blank was also prepared simultaneously by taking 5ml of ferric chloride diluting reagent and 4ml of concentrated sulphuric acid. After 30 mins, the intesity of the colour developed was seen during 540mm filter.

APPENDIX VII

ESTIMATION OF HAEMOGLOBIN BY CYANMETH HAEMOGLOBIN METHOD

Estimation of Haemoglobin by this method was recommended by X International Haematology Congress and WHO Expert Committee on Nutritional Anaemias. This method measures not only Hb but also co-Hb and meth-haemoglobin, except sulpha-Hb.with filter type photoelectric calorimeters. The single relatively broad band of cyan-methhaemoglobin in the green spectral region, has a distinct advantage. This method can be modified to determine Hb in dry blood or filter paper also.

PRINCIPLE

Hb is converted into cyanmeth Hb by the addition of KCN and ferricyanide. The colour of cyanmeth haemoglobin is read in a photo electric colorimeter at 540mm against a standard solution since cyanide has the maximum affinity for Hb; this method estimates the total Hb.

REAGENTS

Drabkin's Diluent solution (Cartweight, 1958).

Sodium bicarbonate - 1g

Potassium Cyanide - 0.05g

Potassium ferricyanide- 0.20g

Distilled water to make one litre.

PROCEDURE

Exactly 5ml of Drabkins solution is measured into a dry test-tube from a burette or a pipette with suction bulb. Blood 20.01ml is transferred (0.02ml) with the help of a haemoglobin pipette into the diluent solution. Usual care in filling and cleaning of loaded Hb pipette must be observed. The pipette is rinsed 3 times with the diluent solution without allowing the formation of air bubbles in the solution.

The blood and the diluent are throughly mixed by rotating the tube.

10 minutes time is allowed for the formation of cyanmethhaemoglobin.

5 ml of diluent solution is used as blank.

The readings are taken in a photo electric colorimeter using 540mm (green filter).

CONSTRUCTION OF A STANDARD CURVE

A std curve can be constructed by using the standard cyan meth Hb solutions (supplied by BDH or V.P. Chest Institute, Delhi).

Standard solution(ml)	Dralkins Diluent(ml)	Hb concn (%)
5	0	100
5	2.5	67
5	5.0	50
5	7.0	40
5	10.0	33

The concentration of the standard is mentioned on the ampoule. The corresponding blood Hb in g/100ml can be obtained by multiplying the concn. on the ampoule by the dilution factor (251).

NOTE

1. Drabkin's solution should be stored in amber coloured bottle. If any precipitate is formed the reagent should be discarded.
2. Since the dilution is a enormous (251 times) accurate measurement of 20ml of blood is absolutely essential for reproductibility. Hb pipettes must be checked for their accuracy by weighing pure mercury upto the mark.

APPENDIX VIII

ESTIMATION OF PCV AND MCHC PERCENTS IN METHOD

PRINCIPLE

The PCV or haemotocrit of blood is determined using heparinised capillary tubes (75x1mm) and micro haemotocrit centrifuge.

PROCEDURE

Blood from the finger tip or collected in EDTA is allowed to run about ½ to 3/4th length of the tube. The tube is sealed on the opposite and using sealing wax or plasticine. The tubes are then transferred to the high speed micro haemotocrit. Centrifuge, and placed in groves of the centrifuge head. They are centrifuged for 5 minutes of 11,000 rpn and read on the reader which gives the direct haematocrit value in volumes present.

MEAN CORPUSCULAR HAEMOGLOBIN CONCENTRATION(MCHC)

$$\text{MCHC \%} = \frac{\text{Haemoglobin}}{\text{PCV}} \times 100$$

APPENDIX IX

ESTIMATION OF BLOOD GLUCOSE - FOLIN WU METHOD

PRINCIPLE

Blood is deproteinized and heated with alkaline copper reagent in Folin-Wu tube and the cuprous oxide formed is treated with acid molybdate solution. When blue colour is obtained the colour is compared with the standard colorimetrically at 600mm.

REAGENTS

1. Sodium tungstate 10%

Dissolved 10g of sodium tungstate in 100ml of distilled water.

2. Phosphomolybdic acid solution:

Dissolved 35g of molybdic acid and 5g of sodium tungstate in 200 ml of water and boiled vigorously for 20-40 minutes to remove ammonia present in molybdic acid; cooled and diluted to about 350ml and added 125ml of concentrated phosphoric acid and made upt the volume to 500ml.

3. Alkaline copper tartarate solution:

Dissolved 46 gms of anhydrous sodium carbonate in about 400ml of water and transferred to a one litre flask. Added 7.5gms of tartaric acid and after it dissolved added 4.5gms of crystalline copper sulphate and made upto 1 litre.

4. Stock standard Glucose:

Dissolved 0.2g of glucose in saturated benzoic acid and made upto 100ml.

5. Working standard:

10ml of the stock standard was diluted to 100ml in a standard flask with distilled water.

6. 2/3N sulphuric acid:

3.2ml acid and 196.31 ml water.

PROCEDURE

Took 3.4ml of distilled water and added 0.2ml of blood and 0.2ml of 2/3N sulphuric acid and then added 0.2ml of 10% sodium tungstate solution. Kept for 20 minutes and then centrifuged. Piepetted out 2ml of the supernatent into 2 Folin-Wu tubes.

0.2ml of standard glucose solution was taken in a series of test-tubes. Added 2ml of alkaline copper tartarate solution and heated to 8 minutes. Added 2ml of phosphomolybdate, cooled and then mixed and made upto 12.5ml with distilled water. The blue colour developed was read in a colorimeter at 600mm.

APPENDIX X

INDIVIDUAL FOOD INTAKE OF MEN VOLUNTEERS

S.No.	Cereals	Pulses	Green-Leafy-Vegeta-bles	Other Vegeta-bles	Roots & Tubers	Milk & Milk Products	Fats & Oils	Sugar & Jaggery	Fruits
1	2	3	4	5	6	7	8	9	10
1.	443	90	20	100	95	400	30	20	103
2.	376	85	15	78	90	450	30	20	50
3.	380	60	30	100	95	400	15	20	20
4.	400	75	50	65	95	350	30	20	40
5.	380	78	10	75	90	400	20	20	100
6.	423	95	25	95	90	400	20	20	20
7.	440	93	20	86	95	350	15	20	25
8.	394	86	20	40	93	400	18	20	56
9.	385	72	30	23	85	400	13	20	55
10.	380	95	33	65	86	375	20	20	44
11.	383	90	20	40	70	400	30	20	77
12.	380	93	22	95	20	380	35	20	89
13.	379	97	25	100	93	383	14	20	20
14.	410	80	50	53	47	200	19	20	26
15.	383	68	39	51	100	400	25	20	50
16.	400	95	35	100	50	400	25	20	55
17.	435	80	20	95	30	400	25	20	55
18.	444	83	22	95	35	400	30	20	80
19.	404	35	20	30	80	400	30	20	100
20.	404	95	20	30	80	400	30	20	100

1	2	3	4	5	6	7	8	9	10
21.	460	95	20	50	20	400	30	20	100
22.	429	97	15	20	100	400	30	20	100
23.	450	35	43	104	25	300	30	20	100
24.	400	89	35	107	25	300	35	20	100
25.	401	45	20	75	25	300	25	20	100
26.	468	65	20	200	25	300	20	20	120
27.	455	30	23	50	65	300	15	20	120
28.	425	75	20	60	85	400	15	20	120
29.	436	80	20	65	85	450	10	20	120
30.	439	82	20	68	89	400	20	20	120
31.	435	90	15	92	60	400	23	20	100
32.	450	90	20	65	60	400	26	20	100
33.	385	86	30	60	65	400	28	20	107
34.	350	45	35	86	69	200	37	20	107
35.	428	48	36	85	30	250	30	20	107
36.	450	91	40	100	25	200	30	20	115
37.	385	94	43	90	25	275	30	20	115
38.	383	75	45	95	20	275	30	20	115
39.	415	72	40	95	75	200	30	20	100
40.	429	75	45	60	60	200	30	20	100
41.	450	75	30	62	77	400	30	20	100
42.	452	61	35	63	79	400	30	20	100
43.	448	66	36	63	30	350	30	20	100
44.	418	60	36	89	45	350	15	20	50
45.	385	47	36	90	30	350	15	20	50
46.	360	47	20	100	30	375	15	20	60
47.	420	60	21	107	15	400	30	20	60
48.	405	60	21	53	75	400	30	20	60
49.	468	65	20	55	75	400	37	20	60
50.	400	95	35	55	85	400	20	20	60

APPENDIX XI

INDIVIDUAL FOOD INTAKE OF WOMEN VOLUNTEERS

S. No.	Cereals	Pulses	Green - Leafy vegetables	Other - Vegetables	Roots & Tubers	Milk & Milk Products	Fats & Oils	Sugar & Jaggery	Fruits
1	2	3	4	5	6	7	8	9	10
1.	349	75	10	30	67	483	38	20	141
2.	366	55	28	54	44	500	30	20	116
3.	385	74	25	52	39	467	27	20	115
4.	467	82	25	61	45	500	30	20	117
5.	353	65	107	23	29	350	28	10	38
6.	284	37	46	18	35	283	24	15	16
7.	270	28	15	33	71	333	12	10	9
8.	369	65	15	51	78	467	35	15	129
9.	394	44	7	26	85	466	35	20	140
10.	382	67	13	54	93	433	37	15	131
11.	355	71	15	25	42	400	26	20	77
12.	417	44	29	30	92	450	38	20	115
13.	373	78	5	46	40	417	15	20	115
14.	404	61	15	22	46	383	32	20	131
15.	401	49	21	32	48	400	15	20	121
16.	332	66	63	35	57	308	16	20	67
17.	196	45	-	24	143	283	20	20	80
18.	383	24	19	54	82	200	17	15	116
19.	313	32	-	20	50	200	17	20	34
20.	348	16	45	20	59	208	12	20	66

1	2	3	4	5	6	7	8	9	10
21.	346	46	15	9	55	200	12	20	21
22.	363	90	50	100	95	200	15	10	30
23.	360	63	100	80	58	275	33	28	47
24.	375	75	57	55	42	250	30	22	33
25.	353	68	50	60	25	330	30	27	83
26.	412	73	3	46	50	383	27	28	30
27.	369	64	22	50	60	416	30	23	40
28.	383	68	57	60	71	400	25	30	107
29.	322	65	57	60	88	400	27	23	100
30.	382	68	57	60	71	200	25	23	100
31.	280	74	10	24	29	350	17	9	40
32.	336	65	38	18	31	350	15	20	65
33.	420	42	25	49	78	350	20	22	69
34.	446	60	15	28	95	383	30	20	33
35.	412	76	44	33	125	357	35	20	107
36.	400	22	32	44	59	308	39	20	116
37.	380	38	59	57	65	205	39	10	67
38.	507	56	53	43	85	200	27	15	100
39.	377	49	64	68	101	200	16	15	115
40.	375	34	66	69	57	265	12	10	131
41.	389	76	18	54	82	200	25	5	8
42.	435	32	41	75	35	250	24	23	15
43.	382	37	55	69	22	400	28	20	29
44.	330	34	32	82	36	389	12	20	27
45.	358	24	46	25	51	200	10	22	32
46.	339	65	85	30	61	285	20	10	50
47.	400	79	67	41	23	300	25	20	33
48.	429	33	23	87	39	200	35	20	45
49.	415	56	101	16	90	250	42	20	37
50.	409	67	49	35	124	200	30	20	20
51.	385	42	54	30	39	200	30	20	16

APPENDIX XII

INDIVIDUAL NUTRIENT INTAKE OF MEN VOLUNTEERS

S. No.	Protein (g)	Energy (Kcal)	Calcium (mg)	Iron (mg)	Vit.A (mcg)	Thiamine (mg)	Riboflavin (mg)	Niacin (mg)	Ascorbic Acid (mg)
1	2	3	4	5	6	7	8	9	10
1.	97.2	3234	1156	45.4	4141	1.70	3.30	16.23	168
2.	76	3676	978	67.2	2691	1.50	2.01	17.52	154
3.	70	2350	1050	75.4	3000	1.02	1.86	16.36	124
4.	98	2678	840	75.4	3015	1.21	1.45	16.43	56
5.	65	2956	750	29.0	1678	1.78	1.21	16.25	57
6.	49	2500	867	58	1589	1.28	1.29	16.86	53
7.	47	2789	877	55	1285	1.27	1.98	17.96	67
8.	54	2657	867	70.6	2085	1.56	1.54	17.01	156
9.	45	2650	474	69	2066	1.09	1.86	17.62	71
10.	63	2616	900	43	2509	1.41	1.01	17.00	76.6
11.	47	2748	925	67	2003	1.46	1.01	16.64	151
12.	76	2420	963	23.5	2446	1.99	1.03	17.01	120
13.	55	2560	925	56.1	7202	1.86	1.06	16.66	118.7
14.	53	2720	1201	57.5	4018	1.54	1.05	16.87	83
15.	50	2600	1147	33.6	2008	1.42	1.02	16.63	56.8
16.	53	2805	1129	60.8	2016	1.62	1.78	16.28	189
17.	53	2626	990	48	2618	1.68	1.05	16.85	114
18.	60	2545	997	21	2068	1.71	1.02	17.65	161
19.	61	2785	568	39	2175	1.79	1.03	17.62	113
20.	52	2766	819	76	2118	1.41	1.09	16.25	121

1	2	3	4	5	6	7	8	9	10
21.	67	2420	902	28.7	2097	1.82	1.01	16.29	121
22.	74	2325	905	52	2248	1.86	1.22	16.00	166
23.	68	2306	906	37	2132	1.92	1.25	17.60	140
24.	78	2778	905	35	2890	1.89	1.28	17.78	186
25.	52	2402	666	25.9	3042	1.71	1.29	16.87	196
26.	51	2300	920	38.5	2089	1.00	1.32	16.07	186
27.	47	2675	987	26.7	3229	1.65	1.33	16.92	132
28.	40	2627	639	25.4	3869	1.86	1.21	16.62	126
29.	50	2509	568	71	3200	1.43	1.05	16.43	58.6
30.	55	2515	879	30.8	2185	1.86	1.00	16.00	55.86
31.	35	2806	817	28.1	1587	1.27	1.06	16.12	57.24
32.	45	2805	595	53	2709	1.45	1.62	17.81	124
33.	63	3012	615	41	3608	1.52	1.20	17.01	124
34.	57	3200	1202	40	3545	1.89	1.02	17.30	115
35.	54	3241	1011	38.5	2078	1.20	1.00	17.48	125
36.	56	2877	1034	30	2105	1.01	1.00	17.56	115
37.	76	2565	989	42	2168	1.86	1.01	17.98	178
38.	77	2644	990	64	2586	1.05	1.05	17.33	~~178.5~~
39.	90	2676	901	66	2687	1.05	1.08	16.33	66.6
40.	95	2727	1180	75	2279	1.21	1.80	17.41	57.2
41.	86	2620	540	67	2108	1.15	1.85	17.65	58.4
42.	48	2905	553	54	2189	1.19	1.62	17.65	56.4
43.	89	2460	545	57	2008	1.29	1.82	17.68	57.2
44.	97	2715	656	52	2000	1.85	1.01	17.42	59.0
45.	61	2527	628	54	2785	1.54	1.06	17.23	113
46.	92	2600	1022	36.1	2805	1.23	1.00	16.10	113
47.	40	2080	1166	34.5	3020	1.52	1.00	16.15	142
48.	59	2690	988	38.0	3115	1.53	1.01	17.51	56.79
49.	87	2079	952	39.0	2950	1.67	1.07	16.00	58
50.	75	2615	943	30	2112	1.05	1.09	16.00	60

APPENDIX XIII

INDIVIDUAL NUTRIENT INTAKE OF WOMEN VOLUNTEERS

S. No.	Protein (g)	Energy (Kcal)	Calcium (mg)	Iron (mg)	Vit.A (mg)	Thiamine (mg)	Riboflavin (mg)	Niacin (mg)	Ascorbic Acid (mg)
1	2	3	4	5	6	7	8	9	10
1.	66.7	2558	895	22.9	1969	1.01	4.38	10.8	46
2.	65.6	2605	826	23.2	1422	1.02	4.59	10.5	69
3.	62.8	2477	762	21.7	923	0.83	3.90	10.6	66
4.	62.1	2199	644	21.8	1090	4.41	1.50	15.4	154
5.	68.0	2490	650	22.8	950	3.22	3.20	12.3	90
6.	65.2	2500	810	25.6	920	4.10	3.00	15.0	68
7.	69.0	2625	860	22.0	1200	2.02	4.69	13.4	74
8.	66.2	2503	743	24.6	1423	4.60	4.98	14.5	85
9.	65.7	2255	725	20.2	1548	4.52	4.82	14.3	83
10.	69.2	2465	623	25.6	1780	4.25	3.01	15.1	76
11.	62.3	2400	813	24.8	1102	2.46	3.82	15.4	68
12.	64.9	2700	750	28.9	1069	3.05	3.65	15.0	65
13.	65.4	2550	810	26.6	1542	3.79	3.48	10.2	68
14.	66.2	2623	805	26.3	1050	4.85	4.15	15.3	66
15.	62.6	2730	833	23.4	1102	3.00	4.18	10.2	64
16.	62.0	2477	846	23.6	960	2.56	4.15	10.21	21
17.	62.6	2585	828	22.9	1103	3.04	3.26	10.75	57
18.	65.2	2500	835	20.7	1240	1.01	4.78	14.15	32
19.	63.9	2525	927	21.3	1179	1.01	4.69	13.75	41
20.	64.8	2602	841	24.9	1260	2.03	4.50	14.01	65

1	2	3	4	5	6	7	8	9	10
21.	66.3	2556	762	24.6	1456	2.65	4.78	15.00	67
22.	66.3	2670	675	24.7	1265	4.30	4.75	10.32	62
23.	65.4	2520	810	25.0	1011	4.23	4.00	15.02	60
24.	62.1	2450	827	22.5	1125	3.42	3.15	12.36	56
25.	61.5	2600	843	23.2	1450	4.10	3.75	14.79	93
26.	66.4	2540	727	23.2	1321	4.15	3.06	13.05	124
27.	69.0	2675	605	25.2	1243	2.17	3.03	10.67	63
28.	63.6	2530	708	22.3	1110	2.75	1.50	12.79	90
29.	64.8	2456	625	21.0	1560	3.39	1.70	13.00	65
30.	64.6	2788	621	21.4	1869	3.22	3.61	14.98	65
31.	63.8	2566	637	25.3	1981	0.96	3.02	15.65	44
32.	65.7	2676	865	22.1	969	4.83	3.32	12.06	68
33.	68.5	2560	873	22.3	950	1.02	3.43	13.79	90
34.	67.6	2550	855	24.3	920	4.40	4.03	14.15	55
35.	62.8	2530	715	25.4	1479	3.25	4.00	14.78	46
36.	65.4	2576	701	22.4	920	2.54	4.15	14.50	20
37.	63.4	2545	760	22.4	876	2.01	4.17	14.39	56
38.	64.5	2455	744	23.2	1169	2.27	4.79	14.40	74
39.	65.4	2400	745	20.1	1105	2.45	4.30	13.25	43
40.	62.3	2450	647	20.3	927	1.01	3.15	13.85	59
41.	66.3	2360	650	22.7	1470	1.01	4.67	13.62	75
42.	65.2	2367	716	21.9	956	1.01	4.86	13.03	64
43.	62.4	2485	715	20.9	975	4.58	3.21	13.52	95
44.	63.5	2574	765	23.4	962	4.79	4.97	12.65	63
45.	62.0	2590	610	23.5	1375	3.37	4.58	13.01	67
46.	62.1	2554	620	25.1	1127	4.68	3.01	14.75	66
47.	65.6	2750	814	20.3	1425	2.46	3.69	14.65	154
48.	63.2	2660	690	23.0	1176	2.35	3.65	14.02	100
49.	64.3	2565	775	20.7	1286	2.30	3.22	13.19	67
50.	62.6	2276	823	24.9	1096	2.32	3.28	12.50	67
51.	65.0	2575	702	20.0	1011	1.00	2.02	12.50	62

APPENDIX XIV

PROFORMA FOR THE ASSESSMENT OF NUTRITIONAL STATUS OF VOLUNTEERS

I. GENERAL INFORMATION

NAME: AGE:

SEX : OCCUPATION:

HEIGHT: ACTIVITY: MILD/MODERATE/ HEAVY

WEIGHT :

II. CLINICAL ASSESSMENT OF VOLUNTEERS

CRITERIA	SCORE
1. GENERAL APPEARANCE	
Emaciated or obese/Moderately well nourished/well nourished	0 / 3 / 5
2. HAIR	
a. Pluckability	0./ 3 / 5
Easy pluckability/slightly brittle	
Normal	
b. Nature:	
Thin and Sparse/Silky and straight/Dense, glistening and normal	

c. Colour

Partial or complete dyspig-
mentation dull and dry/
Normal

3. FACE 0 / 3 / 5

Moon face/Paleness/Normal

4. EYES 0 / 3 / 5

a. Vision

Night blindness/Diminished
Vision/Normal clear vision

CRITERIA

b. Conjunctiva

Wrinkled, thickness and
pigmented/slightly dry on
exposure to sunlight/Normal

c. Bitot spots

Marked/Mild/Absent

d. Cornea

Dryness/Opacity/Normal

5. LIPS 0 / 3 / 5

Angular Stomatitis and/or Cheilosis/
angular Scars and/or fissures,
ulcers/Normal

6. Tongue 0 / 3 / 5

Swollen/red, raw and ulcerated/
Normal

7.	Gums Swollen, spongy, bleeding/receding and loose and/or retracted/Normal.	0 / 3 / 5
8.	Teeth Mottled enamle/caries/Normal	0 / 3 / 5
9.	Glands Thyroid enlargement/Parotid enlargement/Normal	0 / 3 / 5
10.	Skin Dry and desquammation/Pellargrous and/or flaky paint dermatos is/ smooth, elastic and normal	0 / 3 / 5
11.	Nails Koilonychia/lubbed, firm pink and Normal	0 / 3 / 5
12.	Nature of joints Abnornial/Normal	0 / 5
13.	Would Healing Delayed healing/Moderately rapid healing/rapid Healing	0 / 3 / 5
14.	Oedema Present all over the body/present on dependent parts/Absent	0 / 3 / 5

15. Musculature 0 / 3 / 5

Muscle wasting/Moderate/Normal

16. Anaemia 0 / 3 / 5

Severe/Mild or Moderate/Absent

17. Constipation 0 / 3 / 5

Frequent/Sometimes or rare/absent

18. Allergy 0 / 3 / 5

Long term allergy / Short term allergy/absent

19. Acute infection 0 / 3 / 5

Often/Sometimes/Rare

20. History of illness 0 / 3 / 5

6-12 months back/12-24 months/ before three years

MAXIMUM SCORE 100 TOTAL SCORE

NOTE

Evaluation of Criteria (2) and (4)

Normal - 5 points

Presence of any one deficiency symptom - 3 Points

Presence of more than one deficiency symptom - 0 points.

APPENDIX XV

INDIVIDUAL MEASURES OF HEIGHTS AND WEIGHTS OF MEN AND WOMEN VOLUNTEERS

S.No.	Height (cm)		Weight (Kg)	
	Men	Women	Men	Women
1	2	3	4	5
1.	167	158	58	56
2.	165	155	66	48
3.	163	157	65	57
4.	167	157	65	45
5.	165	157	60	46
6.	165	160	60	50
7.	163	158	58	59
8.	158	155	56	59
9.	158	157	65	55
10.	163	157.5	68	50
11.	162.5	157	58	58
12.	158	154	60	56
13.	160	152.5	63	48
14.	160	142.5	65	50
15.	157.5	160	63	47
16.	160	160	62	54
17.	162.5	157.5	60	50
18.	165	155	60	47
19.	162.5	152.5	55	48
20.	155	155	60	48
21.	165	160	56	55
22.	162.5	142.5	68	45
23.	162.5	157.5	62	52
24.	165	157.5	60	49
25.	165	155	60	45

1	2	3	4	5
26.	160	156	59	43
27.	165	157	58	52
28.	165	167.5	60	60
29.	163	155	56	54
30.	163	162.5	57	51
31.	158	150	58	46
32.	160	155	58	49
33.	158	157.5	62	55
34.	172	160	68	59
35.	160	142.5	60	46
36.	160	157.5	65	45
37.	155	152.5	54	45
38.	158	157	57	50
39.	162.5	155	58	48
40.	160	142.5	63	56
41.	155	157	65	52
42.	165	150	65	47
43.	160	155	57	54
44.	160	157.5	63	52
45.	162.5	160	65	58
46.	162.5	155	66	48
47.	167.5	155	61	50
48.	162.5	160	58	52
49.	157.5	150.5	55	47
50.	155	150	56	48
51.	-	160	-	50

APPENDIX XVI

INDIVIDUAL VALUES OF HAEMOGLOBIN, PACKED CELL VOLUME PERCENT AND MEAN CORPUSCULAR HAEMOGLOBIN CONTENT PERCENT OF MEN AND WOMEN VOLUNTEERS

S.No.	Men			Women		
	Haemoglobin (g/dl)	PCV Percent	MCHC Percent	Haemoglobin(g/dl)	PCV percent	MCHC Percent
1	2	3	4	5	6	7
1.	15.8	42	37.6	13.0	39	33.3
2.	16.0	40	40	10.2	32	31.9
3.	15.7	42.2	37.2	13.5	39	34.6
4.	15.6	42	37.1	12.5	36	34.7
5.	15.5	42	36.9	12.4	36.9	33.6
6.	14.6	40	36.5	14.1	32	44.1
7.	14.5	39.9	36.3	11.5	33	34.9
8.	14.0	39.9	35.8	14.1	32	44.1
9.	14.6	40	36.5	12.8	36.6	36.9
10.	14.6	39.9	36.6	13.8	39	35.4
11.	16.2	40.1	40.4	14.0	32	44.1
12.	16.0	40	40	14.0	32	44.1
13.	15.5	40	38.6	14.4	34	42.4
14.	14.8	40	37	11.1	34	32.7
15.	14.5	39.9	36.3	13.4	39	34.4
16.	14	39	35.9	12.5	34.8	35.92
17.	14	40	35	12.5	34.8	35.9
18.	16.0	42	38.1	12.6	34	37.1
19.	14.5	40	36.1	13.8	38.2	36.1
20.	14.5	40.2	36.1	11.5	35	32.9
21.	14	39	35.9	13.4	38	35.3
22.	15.2	38.8	39.2	14.0	34	41.2
23.	14.8	41	33.6	13.4	36	37.2
24.	14	39	35.9	10.2	32	31.9
25.	14.2	42	33.8	10.4	32.2	32.8

1	2	3	4	5	6	7
26.	14.4	40	36	10.9	33	33.0
27.	14	41	34.2	10.4	32	32.5
28.	14.6	42	34.8	10.4	32	32.5
29.	14.2	40	35.5	11.7	35	33.4
30.	14.7	42	35	12.6	37	33.2
31.	14.6	40	36.5	11.5	34	33.8
32.	14.0	39	35.9	10.5	33	31.8
33.	14.8	42	35.2	14.6	36	40.6
34.	13.8	34	40.6	14.6	36	40.6
35.	13.1	38.5	34	10.8	32	33.8
36.	13.6	39.7	34.2	12.2	33	36.2
37.	14.5	42	34.5	10.8	34	31.8
38.	15.6	39	40	10.5	32.2	32.6
39.	14.6	42	34.8	13.7	35	39.1
40.	15.5	42	36.9	13.4	38	35.3
41.	16.0	48	33.3	11.5	36	31.9
42.	14.8	40.1	36.9	11.1	36	30.8
43.	15.2	40	38	13.7	38	36.1
44.	14.5	39.9	36.3	11.5	38	30.3
45.	14.0	40	35	10.8	32	33.8
46.	16.3	45	36.7	10.2	32	31.9
47.	14.8	40	37	11.9	36	33.1
48.	14.2	39.9	35.6	12.5	36	34.7
49.	15.0	40	37.5	10.9	34	32.1
50.	14.4	40	36.0	11.7	36	32.5
51.	-	-	-	10.2	32	31

APPENDIX XVII

INDIVIDUAL VALUES OF SERUM IRON, TOTAL IRON BINDING CAPACITY AND PERCENT SATURATION LEVELS FOR MEN AND WOMEN VOLUNENTEERS

S.No.	Men				Women			
	Serum Iron mcg/dl	TIBC mcg/dl	UIBC mcg/dl	Percent saturation Transferrin	Serum Iron mcg/dl	TIBC mcg/dl	UIBC mcg/dl	Percent saturation Transferrin
1	2	3	4	5	6	7	8	9
1.	66.2	202.7	136.5	32.7	56.6	153.1	86.5	37.0
2.	56.5	153.1	96.5	37.0	59.0	166.1	107.1	35.5
3.	43.8	222	178.2	19.7	65	293.1	228.1	22.2
4.	88	287.6	199.6	30.6	89.2	280	190.8	31.9
5.	56.5	153.6	97.1	36.7	105	340.5	235	30.8
6.	110.8	298	187.8	36.9	112	310	198	36.1
7.	164.2	330.4	166.2	49.6	101.1	294.7	193.6	34.3
8.	155.5	346.4	190.9	44.9	100.5	299.3	198.8	33.5
9.	103.2	296.7	193.5	34.7	159.2	335	175.8	46.5
10.	120	325	205	36.9	68.5	269	200.5	25.5
11.	99	289	190	34.2	142	305	163	46.5
12.	134	328	194	40.8	58.7	210	151.3	28.1
13.	150	344	194	43.6	147	268	121	54.9
14.	128.7	285.6	156.9	45.1	112.9	279	166.1	40.4
15.	139.2	315	175.8	44.2	175	280	105	62.5
16.	110.5	289	178.5	38.2	122	280	158	43.6
17.	118.1	320	201.9	36.9	132.4	337	204.6	39.3
18.	100.2	313.9	213.7	31.9	90.5	280	189.5	32.3
19.	97.7	281	183.3	34.8	121.3	330.3	199	36.7
20.	117.4	300	182.6	39.1	109	310	202	35.1
21.	107.8	299.1	191.3	36.0	95	279	184	34.1
22.	168	360	192	46.6	101.4	258	156.6	39.3
23.	95	259	164	36.7	118	310	192	38
24.	152.3	348	195.7	43.7	97.5	298	200.5	32.7
25.	104.9	283.6	178.7	36.9	99	298	189	33.2

1	2	3	4	5	6	7	8	9
26.	143	322	179	44.4	103.7	298	194.3	34.7
27.	151	310	159	48.3	98	287	189	34.1
28.	109.8	296	186.2	37.1	85	268	183	31.7
29.	59.5	178.2	118.7	33.9	147	344	197	42.7
30.	162.2	346.4	184.2	46.8	115	290.5	165.5	40.9
31.	118.1	281.2	163.1	41.9	120	306	186	39.2
32.	89.9	289.6	199.7	31.0	114	309	195	36.9
33.	106.7	298.2	191.5	35.8	120	310	190	38.7
34.	146.3	300.5	154.2	48.7	101.2	285	183.8	35.5
35.	116.2	275	158.8	42.2	100.4	268	167.6	37.5
36.	113.4	282.1	168.7	40.2	97.3	292	194.7	33.3
37.	90	280	190	32.1	103	322	219	31.9
38.	68.9	165.3	96.4	41.7	86.7	308	211.3	31.4
39.	117.7	279	181.3	39.4	104.3	296	191.7	35.2
40.	118	286	168	41.2	111.6	303	191.4	36.8
41.	131.5	326	194.5	40.3	98.3	296	197.7	33.2
42.	115.3	278	162.7	41.5	116.1	303	186.9	38.5
43.	112.9	301	187.9	37.5	106.4	296	187.6	35.9
44.	154	310	156	49.7	121.3	340	218.7	35.7
45.	99.3	287	187.7	34.6	111.3	310	198.7	35.9
46.	165	335	170	49.3	125	344	219	36.3
47.	168	365	197	46.0	86	285	189	30.2
48.	149.7	330	180.3	45	146	303	157	40.2
49.	126.2	316.2	190	39.9	110.5	281	170.5	39.3
50.	131.2	350	218.8	37.5	105	340.5	235	30.8
51.	-	-	-	-	115	348	233	33.0

APPENDIX XVIII

INDIVIDUAL VALUES OF TOTAL SERUM PROTEIN, ALBUMIN AND GLOBULIN OF MEN AND WOMEN VOLUNTEERS

S.No.	Men			Women		
	Total Serum Protein g/dl	Albumin g/dl	Globulin g/dl	Total Serum Protein g/dl	Albumin g/dl	Globulin g/dl
	1	2	3	4	5	6
1.	7.4	4.7	2.7	7.6	4.09	3.69
2.	6.3	3.9	2.4	7.6	5.48	2.36
3.	6.8	5.3	1.5	5.9	3.6	2.3
4.	7.6	4.3	3.3	7.2	4.3	2.9
5.	6.5	4.2	2.3	6.8	4.3	2.5
6.	7.3	5.3	2.0	6.6	3.9	2.7
7.	6.3	3.9	2.4	7.1	4.0	2.1
8.	7.7	4.2	3.5	7.7	5.2	2.6
9.	6.9	4.1	2.8	7.0	5.0	2.0
10.	7.5	4.7	2.8	6.5	4.3	2.2
11.	6.2	3.9	2.3	6.5	4.3	2.2
12.	5.9	3.6	2.3	6.7	4.2	2.5
13.	7.1	4.3	2.8	6.6	4.5	2.1
14.	7.2	4.2	3.0	4.95	3.5	2.45
15.	6.1	4.3	2.8	7.2	4.9	2.3
16.	6.6	4.25	2.35	8.0	6.1	1.9
17.	6.8	4.8	3.0	6.8	4.2	2.6
18.	7.5	5.5	2.0	6.6	3.9	2.7
19.	7.8	5.2	2.6	6.7	4.8	1.9
20.	6.7	4.4	2.3	7.6	4.5	3.1
21.	7.2	4.1	3.1	7.6	4.9	2.7
22.	7.2	4.8	2.4	5.9	3.7	2.2
23.	7.3	4.3	3.0	6.1	3.9	2.2
24.	7.6	5.2	2.4	6.3	4.6	1.7
25.	7.5	4.6	2.9	6.6	5.1	1.5

	1	2	3	4	5	6
26.	7.3	4.6	2.7	6.4	4.7	1.7
27.	6.4	4.3	2.1	5.8	3.6	2.2
28.	6.5	4.1	2.4	7.2	5.9	1.3
29.	7.5	4.7	2.8	6.5	4.5	2.0
30.	7.7	4.2	3.5	6.7	4.8	1.9
31.	6.5	4.2	2.3	6.4	5.1	1.3
32.	7.2	4.5	2.7	7.1	4.5	2.6
33.	6.2	4.0	2.2	6.2	4.8	1.4
34.	5.9	3.8	2.1	7.2	5.2	2.0
35.	7.0	4.0	3	7.2	4.8	2.4
36.	7.7	5.8	1.9	6.5	4.8	1.7
37.	6.5	4.3	2.2	7.6	5.1	2.5
38.	7.3	5.0	2.3	6.8	4.9	1.9
39.	7.6	5.1	2.5	6.5	4.5	2.0
40.	7.2	4.5	2.7	7.1	5.3	1.8
41.	6.9	4.5	2.4	7.4	5.8	1.6
42.	6.5	4.3	2.2	6.8	4.5	2.3
43.	6.8	4.9	1.9	6.6	4.9	1.7
44.	7.5	4.7	2.8	5.3	4.8	1.5
45.	6.5	3.8	2.7	6.2	4.9	1.3
46.	5.9	3.9	2.0	6.1	4.3	1.8
47.	7.2	4.5	2.7	5.9	3.8	2.1
48.	7.5	4.6	2.9	7.3	5.1	2.2
49.	5.8	3.5	2.3	7.2	4.8	2.4
50.	7.6	4.7	2.0	6.6	4.6	2.0
51.	-	-	-	6.8	4.8	2.0

APPENDIX XIX

INDIVIDUAL VALUES OF SERUM RETINOL AND SERUM CALCIUM OF MEN AND WOMEN VOLUNTEERS

S.No.	Men		Women	
	Serum Calcium (mcg/dl)	Serum Retinol (mg/dl)	Serum Calcium (mcg/dl)	Serum Retinol (mg/dl)
1	2	3	4	5
1.	39.1	8.9	41.3	11.5
2.	32.0	10.8	36.5	9.5
3.	38.2	10.9	32.1	9.8
4.	43.2	10.0	38.0	9.7
5.	33	10.5	40	10.6
6.	40.7	10.3	40.6	10.0
7.	46.9	10.0	47.0	10.7
8.	47.8	10.0	28.6	10.4
9.	31.5	9.8	30.0	10.2
10.	54.6	9.0	32.9	9.6
11.	30.4	9.9	37.2	10.2
12.	29.9	9.9	33.4	10.3
13.	37.1	10.6	44.0	11.6
14.	4.5.3	10.0	44.9	11.9
15.	41.6	10.3	32.3	11.0
16.	38.0	11.0	36.7	11.5
17.	51.2	11.5	30.3	11.6
18.	40.3	11.3	36.0	11.6
19.	39.0	10.8	35.1	11.0
20.	37.0	9.9	32.5	11.0
21.	29.8	10.0	44.4	11.2
22.	36.3	9.1	42.6	11.5
23.	42.7	9.8	42.1	11.1
24.	40.5	10.4	31.8	11.4
25.	42.1	10.8	37.5	11.0

1	2	3	4	5
26.	42.5	11.5	42.0	11.2
27.	49.0	11.2	44.6	11.2
28.	46.9	11.0	33.9	11.0
29.	33.3	11.0	29.7	10.1
30.	41.2	11.3	38.0	10.0
31.	36.0	10.5	48.0	10.0
32.	36.0	10.2	52.4	9.9
33.	27.0	11.6	50.4	9.8
34.	35.6	10.6	53.7	10.7
35.	38.3	10.4	32.5	10.3
36.	40.7	10.3	36.2	10.0
37.	48.6	10.8	36.0	10.5
38.	54.5	10.8	38.0	10.0
39.	35.2	10.0	38.0	10.0
40.	42.1	9.8	40.0	11.0
41.	47.3	9.1	42.7	11.7
42.	44.0	9.9	35.6	10.8
43.	41.5	9.6	36.8	10.0
44.	51.0	10.9	39.9	11.3
45.	39.5	10.0	37.5	11.0
46.	50.0	10.8	38.6	11.0
47.	44.0	10.3	36.5	10.5
48.	38.0	10.4	35.1	10.2
49.	28.7	10.7	32.3	10.7
50.	40.0	9.7	44.0	10.5
51.	-	-	32.0	10.9

APPENDIX XX

INDIVIDUAL VALUES OF SERUM COPPER AND ZINC OF MEN AND WOMEN VOLUNTEERS

S.No.	Men		Women	
	Serum Zinc mcg/dl	Serum Copper mcg/dl	Serum Zinc mcg/dl	Serum Copper mcg/dl
1	2	3	4	5
1.	109	120	107	79
2-	99	104	65	98
3.	80	96	78	95
4.	90	100	100	77
5.	104	102	94	82
6.	100	102	94	81
7.	100	122	96	100
8.	100	134	102	111
9.	67	86	100	99
10.	102	85	86	102
11.	94	98	88	109
12.	99	99	90	111
13.	117	100	100	121
14.	109	112	107	94
15.	98	101	102	98
16.	92	106	101	101
17.	76	110	115	115
18.	85	98	120	120
19.	100	87	102	125
20.	102	89	60	130
21.	98	135	104	79
22.	105	111	91	110
23.	107	102	102	105
24.	100	101	102	107
25.	102	108	102	100

1	2	3	4	5
26.	104	109	120	131
27.	100	100	126	131
28.	100	106	96	130
29.	76	97	60	99
30.	90	98	65	76
31.	93	100	78	77
32.	100	100	93	75
33.	78	100	102	74
34.	85	92	98	72
35.	83	89	98	90
36.	80	82	90	90
37.	96	79	111	95
38.	119	132	94	75
39.	111	122	94	98
40.	75	118	98	108
41.	115	92	90	110
42.	100	95	69	112
43.	94	129	68	85
44.	100	130	77	93
45.	76	90	95	87
46.	92	99	90	87
47.	100	102	100	94
48.	99	102	86	120
49.	101	96	68	125
50.	84	135	81	112
51.	75	137	-	-

APPENDIX XXI

INDIVIDUAL VALUES OF BLOOD GLUCOSE AND SERUM CHOLESTEROL OF THE VOLUNTEERS

S.NO	Men		Women	
	Blood glucose mg/dl	Serum cholesteral mg/dl	Blood glucose mg/dl	Serum cholesteral mg/dl
1	2	3	4	5
1.	90	173.3	86	164.1
2.	95	173.9	88	134.2
3.	108	178.9	90	150.5
4.	106	155	100	168.9
5.	100	135	119	96.5
6.	118	160	120	104
7.	85	189	87	125
8.	96	145	89	220
9.	87	185	80	205
10.	87	160	100	210
11.	80	190	106	200
12.	79	165	108	185
13.	112	165	90	175.3
14.	120	178	90	173.3
15.	105	185	90	176
16.	106	125	95	135
17.	106	133	90	186.5
18.	108	135	78	175
19.	110	175	80	165
20.	111	186	86	163.5
21.	85	200	104	130
22.	90	220	90	185
23.	92	190	96	230

1	2	3	4	5
24.	95	212	94	140
25.	100	139	98	165
26.	120	130	100	150
27.	115	125	110	135
28.	88	185	102.6	225
29.	98	200	105	216
30.	86	205	112	155
31.	88	150	105	155.3
32.	100	130	106	205
33.	101	160	105	220
34.	110	185	100	130
35.	115	189	98	145
36.	120	179	84	118
37.	98	173.5	76	120
38.	99	173.9	75	150
39.	100	148	88	165
40.	100	190	104	200
41.	100	220	120	118
42.	95	230	75	200
43.	88	210	74	200
44.	96	205	80	210
45.	80	205	85	240
46.	76	170	95	204
47.	104	189	110	135
48.	110	165	96	175
49.	85	238	98	150
50.	82	229	100	150.5
51.	-	-	88	165

APPENDIX-XXII

't' VALUES OF ESTIMATED NUTRIENTS

Sample size = 101

Formula For calculation of 't':-

$$'t' = \frac{\text{Difference}}{\text{Standard Error (S.E)}}$$

$$= \frac{\text{Difference between the means of sample}}{\text{Standard Error (S.E)}}$$

$$\text{i.e. } 't' = \frac{\bar{X}_1 - \bar{X}_2}{\sqrt{\frac{\sigma_1^2}{n_1} + \frac{\sigma_2^2}{n_2}}}$$

Where,

n_1 and n_2 = Total no. of samples in each group.

$\bar{X}_1$ and $\bar{X}_2$ = Mean value of the two groups in comparison respectively.

σ_1 and σ_2 = Standard deviation of the two groups in comparison respectively.

Here, n, $\bar{X}_1$ and σ_1 = Total numbers, mean and standard deviation of men volunteers

n_1, $\bar{X}_2$ and σ_2 = Total number, mean and standard deviation of women volunteers.

S.No.	Parameters	$\bar{X}_1$	1	$\bar{X}_2$	2	S.E	't'value
1.	Haemoglobin	14.8	0.74	12.2	1.4	0.22	9.2
2.	PCV	40.5	1.9	34.7	2.3	0.41	6.5
3.	MCHC	36.5	1.8	35.1	3.7	0.58	2.4
4.	Serum Iron	117	30.9	107.8	23.6	5.4	1.3
5.	TIBC	293	49.1	294	37.8	8.7	0.1
6.	UIBC	176	27.1	186	29.9	5.7	1.76
7.	Percent Transferrin satutation	39.5	5.9	36.5	6.4	1.2	2.4
8.	Total Protein	6.95	0.57	6.78	0.54	0.1	1.5
9.	Serum Albumin	5.1	1.23	4.6	0.56	0.2	2.63
10.	Serum Globulin	2.5	0.42	2.2	0.50	0.09	3.3
11.	Serum Retinol	40.58	6.99	38.28	5.8	1.2	1.7
12.	Serum Calcium	10.36	0.64	10.69	0.62	0.07	2.6
13.	Serum Zinc	95.7	11.5	93.1	14.9	2.6	0.96
14.	Serum Copper	104	13.9	99.7	16.4	3.0	1.42
15.	Blood Glucose	98.5	11.7	95.01	11.7	2.3	1.49
16.	Serum Cholesterol	176.8	28.68	170.6	34.8	6.3	0.96