Serum Zinc, Selenium and Chromium Levels in Normal Persons

BY

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Introduction

I. INTRODUCTION

Of the large number of chemical elements found in the human body only a few have demonstrable biochemical or physiological functions. These elements can be considered in five groups. The first includes carbon, Hydrogen, Oxygen, nitrogen and sulfur, the major components of the body molecules. The second group includes the nutritionally important minerals calcium, phosphorus, magnesium, sodium potassium and chloride. The third group includes the trace elements chromium, cobalt, copper, zinc, iron, manganese molybdenum and selenium. A fourth group contains additional elements required for animal nutrition but having no known essential functions in humans; arsenic, cadmium, nickel, silicon, tin and vanadium. The final group contains elements such as mercury and lead that are clearly toxic(Martin et al.,1985). Among the trace elements the range of requirement runs from milligram quantities for zinc, iron manganese down to microgram quantities for some of the newly discovered micronutrient elements such as chromium and selenium. A combined total of only about 25 to 30 g of all trace elements (about an ounce) exists in the human body (compared with over 1000g of calcium alone (Jean Bogert et al.,1983). An element is considered by mertz (1978) to be essential

if its deficiency consistently results in impairment of a function from optimal to suboptimal.

The only property that the essential trace elements have in common is that they normally occur and function in living tissues in low concentrations. The characteristic concentrations and functional forms of the trace elements must be maintained within normal limits if the functional and structural integrity of the tissues is to be safeguarded and the growth, health and fertility of the animals are to remain unimpaired. The trace elements act primarily as catalysts in enzyme systems in the cells, where they serve a wide range of functions. In this respect their roles range from weak ionic effects to highly specific associations known as metalloenzymes. In the metalloenzymes the metal is firmly associated with the protein and there is a fixed number of metal atoms per molecule of protein. These atoms cannot be replaced by any other metal (Eric J.Underwood, 1977). However, vallee (1978) has shown that cobalt and cadmium can be substituted for the native zinc atoms in several zinc enzymes to remain active.

Trace amounts of zinc are present in all living matter. Approximately 263mg are distributed throughout the body (Roslyn et al., 1980). Studies in humans presently indicates that zinc is needed for achievement of normal

growth, for normal sexual maturation and function, for the maintenance of a normal appetite and taste avity, cell division, wound healing disease resistance, healthy skin and even sharp night vision (Walravents, 1979).

Over eighty enzymes are known to require zinc as part of their prosthetic group. They include alcohol dehydrogenase, carbonicanhydrase, Alkaline phosphatase, carboxy peptidase A and B, DNA and RNA polymerases, certain collagenases and enzymes involved in Vitamin-A metabolism. Zinc is found in high concentration in the prostate glands, sperm cells and eyes where it presumably plays important but still unknown function (Lehninger,1984). Zinc is probably also required for the synthesis of protein, RNA and DNA (Slater et al.,1975). It also appears necessary for bone development (Coclhoun et al., 1974). Zinc is also present in the pancreatic islet cells associated with insulin production and the combination of insulin with zinc increases the duration of insulin activity (Varley Gowenlock and Bell, 1980).

Low levels of serum zinc have been found in persons with alcoholic liver diseases, tuberculosis, and in women who are pregnant or taking contraceptives, indicative of borderline intakes, malabsorption syndrome, pancreatitis, diabetes, sickle cell anaemia (John et al., 1978).

The clinical signs of zinc deficiency occur if the level of zinc is below 5-6 μ mol/litre (Weismann,1984). General effects include impaired growth, diarrhoea, slow wound healing and apathy. Cutaneous signs are best exemplified in acrodermatitis enteropathica (AE) which is due to zinc deficiency. There is loss of hair, with redness and crusting particularly round the mouth and in the urogenital area. A secondary deficiency of zinc, induced by high intakes of calcium, phosphorus, and phytate is associated with high incidence of cataract in young salmon. Zinc supplementation successfully prevented cataract appearance and also improved growth (Richardson, et al.,1985).

Selenium-The double duty protector. The same mineral that seems to block certain types of cancer now looks like a guardian of heart health as well (John Feltman,1979). Gerhard N Schrauzer(1979) describes the exciting new evidence that selenium, an important dietary trace element, plays a vital role in maintaining good health. The emerging possibility is that this late-blossoming nutrient helps to prevent the twin scourges of our modern civilization: Cancer and heart disease. Selenium level, was positively associated in both sexes with cancers of liver and stomach with Hodgkins disease and Leukemia (Schrawzer et al.,1979).

Selenium has been shown to be an essential trace element necessary for the enzymatic activity of glutathione peroxidase (GSHPX). Human blood selenium concentration is variable and depends in part on the selenium content and bioavailability in foods and water consumed. Population consuming mainly plant products may have blood selenium levels that differ from population that consume fish and meat products (Shultz T.D., and Leklem E.J.,1983).

An excess of polyunsaturated fatty acids is a specific antagonist to selenium in our body metabolism. Once the vegetable oil content rose above 2 to 3 % of the total diet creates selenium deficiency, diseases like cystic fibrosis, muscular dystrophy, multiple sclerosis sudden infant death syndrome, Kwashiorkor and hyaline membrane diseases were observed in sheeps, pigs, cattles, horses and chickens cystic fibrosis in humans could be a result of zinc deficiency (Marilyn et al.,1986).

Chromium is an essential trace element of both man and animals (Vanrij and Mckenzie J.M.,1979). Although chromium as a component of several enzyme systems, may be important in nucleic acid metabolism, its physiological role remains unclear. Chromium appears to play a role in potentiating the action of insulin and a trivalent chromium nicotinic acid complex has been referred to as "Glucose tolerance cofactor". (John Bernard Henry,1986). It may also be possible that chromium has a role in

lipid metabolism (Mertz. W,1981). Tissue chromium levels have been found by some workers to decline with age whether this is a normal aging process of a reflection of long term inadequate intake is not known (Bunker et. al., 1984).

It is not certain whether chromium deficiency is a factor in the etiology of diabetes or merely a consequence of the disease, since chromium supplementation has not been shown to be beneficial in adult diabetes (John Bernard Henry, 1986). Chromium deficiency condition has also been described in pregnancy, diabetes and in protein calorie malnutrition(Gurson and Saner, 1982).

Chromium excretion in urine is dependent not only on the chromium stores but also in some degree on the amount excreted. The amounts of excreted chromium should be expressed on the basis of creatinine excretion otherwise adjustments for urinary volume have to be made (Joseph Hubert and Dennis Shapcott, 1979). In rats fed diets deficient in chromium and proteins the capacity for the incorporation of certain aminoacids in heart muscle was reduced (Swaminathan,1982).

In view of the importance of zinc, selenium and chromium in maintaining health it is of interest to study their levels in serum of normal persons with different food habits.

Review of Literature

II. REVIEW OF LITERATURE

The review of literature pertaining to the study "Serum Zinc, Selenium and Chromium levels in normal persons" is discussed under the following headings:-

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4. CHROMIUM TOXICITY

A. ZINC:

1. SOURCES OF ZINC:

(i) Human Food Stuffs:-

Human Food Stuffs	Zinc (mg/100g)
Rich sources:	
Herring and oysters	70-150
Good Sources:	
Cereals and millets	2-6
Pulses, oilseeds and nuts	3-6
Liver	3-10
Meat, Fish and eggs	3-8

Zinc occurs in fair amounts in common cereals, pulses and vegetables (Swaminathan, 1983). Variation in zinc content is high within types of foods, as well as among the different class of foods, due to the effects of soil type and fertilizer treatment (E.Underwood, 1977).

(ii) Animal feeds and fodders:

The zinc concentration in plants usually falls with advancing maturity and leguminous plants invariably carry higher zinc levels than grasses grown and sampled under the same conditions (Lang et al., 1972). Heavy dressings with lime and to a lesser

extent with superphosfate can greatly reduce pasture zinc levels (Anonymous, 1977).

The cereal grains used as the basis of pig and poultry rations typically contain 20-30 ppm zinc, with appreciably higher levels in most materials used as protein supplements. Typical values for soybean, peanuts and linseed meals may be given as 50-70 ppm zinc. The zinc content of fish meal, whale meal and meat meal are normally much higher than that of soybean meal (Lunde et al., 1977).

2. OCCURRENCE OF ZINC IN TISSUES AND FLUIDS:

(1) General distribution:

The body contains an average of about 1.8g zinc. In man the prostate is the organ with the highest concentration of zinc with liver, kidney and muscle containing about half as much and heart about a third (Varley et al., 1980). The mean zinc concentration of human dental enamel is reported to be 203± 12µg/g dry weight (Losee et al., 1979).

T A B L E II

Typical zinc concentration of Normal Soft Tissues in mcg/g Fresh tissue

	Tissue	Man[b]	Man[c]	Monkey[d]	Rat[e]	Pig[f]
1.	Adrenal	12	-	16	-	33
2.	Brain	14	13	-	18	-
3.	Heart	33	-	22	21	-
4.	Kidney	55	37	29	23	40
5.	Liver	55	76	51	30	40
6.	Lung	15	14	19	22	-
7.	Muscle	54	39	24	13	-
8.	Pancreas	29	-	48	33	45
9.	Prostate	102	-	-	223	-
10.	Spleen	21	-	21	24	28
11.	Testis	17	13	17	22	-

[b]Tipton and Cook (1973)

[c]Hamilton et al., (1977)

[d]Macapinlac et al.,(1976)

[e]Gilbert and Taylor (1977)

[f]W.G.Hoekstra, Private Communication.

The heart and the testis remain relatively insensitive to very high zinc intakes, but large increase in the zinc concentration of the plasma, liver, kidney, and spleen have been demonstrated in rats fed a normal diet plus 1000 and 2000ppm of supplementry zinc for 15 days (Chen et al., 1977). Comparable large increase in tissue zinc, other than in the heart and muscle, have been observed in calves at high intakes following the breakdown of zinc homeostasis (Stake et al., 1979)

(ii) Eye Tissue:

The eyes would be a sorry sight without zinc. The main circulatory layer of the eye, the choroid contains more zinc than any other part of the body. The retina, a tissue that helps relay visual impulses to the brain contains high levels of zinc. Eye tissues other than the choroid and the iris carry more normal zinc concentrations (Billogottlieb, 1979).

(iii) Male Sex Organs and secretions:

Higher zinc concentrations occur in the prostate gland of the rat, rabbit and man and in the human

seminal fluid and spermatozoa. The following average values may be given; Human prostate 859; dorsolateral rat prostate 891; human semen, first fraction of ejaculate, 2930, second fraction 1400, third fraction 910 and human spermatozoa, 1990 ppm zinc sperm rich boar semen is intermediate between bull semen (10mcg/ml) and human semen (50-200mcg/zn/ml) (Prokaska et al., 1977).

(iv) Nails and hair:

The levels of zinc in the nails of human subjects is reported by Smith to range from 93 to 292 ppm and to average 151 ppm. The head hair of human subjects ranged similarly from 92 to 255 ppm and averaged 173 ppm zinc (Schroeder et al., 1979). In a group of normal pregnant women a small fall in hair zinc levels in a late pregnancy has been observed (Hambidge et al.,1978)

(v) Zinc in blood:

(a) Forms and distribution: 30-40% of plasma zinc firmly bound to a specific α_2 macroglobulin and the remainder is more loosely bound to albumin (Parisi and Vallee, 1978). Over 75% of whole blood zinc is contained in red cells, mainly associated with carbonicanhydrase (Dickerson et al., 1978). 75-88% of the total zinc of normal human blood is contained in the red cells, 12-22% in the plasma and 3% in the leukocytes (Vallee and Gibson, 1977). Human blood platelets contain zinc

in amounts ranging from 0.2 to 0.45mcg/10^9 platelets with a mean of 7.1 mcg of platelet zinc in 100 ml of whole blood (Foley et al., 1976). The zinc concentration of the human serum is consistantly higher than that of plasma by an average of 16% of this increase, 44% is shown to be derived from platelets disintegrating during clotting, 39% from slightly greater dilution in plasma, and 4.0% from hemolysis (Halsted and Smith, 1978)

b) b) Normal levels: The normal levels of zinc in serum is 100(50-150 mcg/dl) (Tierney et al., 1986). Diurnal variations were slight, with a small circadian rythm related to the ingestion of food. Plasma zinc returns to the fasting levels within three hours of taking food (Burr, 1978). Henkin et al., 1977) obtained a slightly lower mean of 48 ± 3 mcg of zinc/dl serum for women at normal delivery compared with a mean of 83 ± 3 mcg zn/dl for serum from the umbilicalcord of their infants.

In a study of normal infants, total plasma zinc concentrations are found to be at adult levels in the new born. These values fall to just below adult levels within the first week of life, fall further at 2-3 months of age, return toward adult values at 4 months and except for a fall at about 1 year of age remain at adult levels throughout the remainder of infancy

(Hankin et al., 1978).

(vi) Zinc in human milk:

The zinc concentration of human milk is appreciably lower than that of cow's milk, although individual variability is high (Picciano et al., 1973).

(vii) Zinc in Avian Egg:

The zinc content of eggs from hens consuming a good laying ration has been reported to average 762±11.4 mcg (Dewar, et al., 1974). Lower levels of zinc than those just given occur in eggs from hens on zinc deficient diets.

3. THE BIOCHEMISTRY OF ZINC:

(i) Absorption:

Dietary zinc intake is normally 0.15-0.23 mmol daily of which two-thirds is absorbed, all through the small intestine. Intestinal mucosa contains two zinc-binding proteins and zinc absorption may be inversely related to intestinal mucosal zinc content. Zinc and copper inhibit absorption of each other, suggesting that they may share certain aspects of absorption. In man absorption of radioactive zinc is apparent fifteen minutes after ingestion and peak plasma levels are reached in four hours (Chambertain et al., 1984). phytate can bind Ca 2+ Mg 2+ and Zn^{2+} very tightly at the multiple phosphate groups preventing these essential metal ions from being absorbed. Phytates are found only in plant

foods especially grains (Lehninger, 1984).

(ii) EXCRETION:

Zinc leaves the body largely by way of the feces. Fecal zinc consists mostly unabsorbed dietary zinc with small amount of endogenous orgin secreted into the small intestine. The quantity of zinc excreted in the urine of healthy human adults is small (0.3-0.6 mg/day), compared with the 10-15mg/day normally ingested. The amounts so excreted do not vary greatly with dietary zinc levels and are not significantly increased following zinc ingestions (Robinson, et al.,1978).

Urinary zinc excretion is well above normal in nephrosis, postalcoholic hepatic porphyria, hyperthyroid patients(Bremmer and Fell 1977). Despite the increased urinary loss, the plasma zinc level remain normal in hyperthyroid patients and there is no correlation between plasma and urine zinc level. One possible explanation for the increased urinary zinc loss in thyrotoxic patients might be an increase in serum aminoacids and other low molecular weight metabolites capable of complexing with zinc and making it ultrafilterable (Aihara et al., 1984).

Significant quantities of zinc can be lost in the sweat, espicially in the tropics. Prasad et al., (1986) report that sweat of normal individuals average 1.15 $\pm$ 0.30 mcg zinc /ml. Most of this is present in

the aqueous phase, ie., not associated with the cellular elements as occurs with iron. In zinc deficient patients the mean zinc level in sweat is reduced to 0.6 ± 0.27 mcg/ml.

Menstrual losses of zinc are small and appear to be of little nutritional significance. If the zinc level of whole human blood is taken as 9 mcg/ml and an average volume of menstrual flow as 50ml, a loss of 450mcg zinc per period can be estimated. This represents only 15 mcg zinc/day dietary in take (Schrader et al., 1975).

(iii) Functions and Effects of zinc deficiency:

a) Growth, Appetite and Taste: Zinc is important in man for growth and sexual development (Varley Gowenlock and Bell, 1980). Dietary deficiency occurs in vegetarians eating predominantly wheat flour possibly due to phytate binding of zinc preventing absorption. Affected males are dwarfed and showed delayed puberty, both features being specifically reversed by zinc supplements.

The growth inhibition of zinc deficiency results partly from impaired appetite. ie. reduced food consumption and partly impaired food utilization. It is seen that poor growth and appetite, together with hypogeusia,

in young children in Denvar is associated with sub-normal hair zinc levels (Hambidge et al., 1979).

Zinc deficiency can wither the taste buds and blocks sense of smell. In acute deficiency smell and taste acutally become unpleasant. And in the past few years, many studies have shown that when people who have lost some or all of their sense of taste take zinc supplements their sense of taste or taste avity improves (Billogottlieb, 1979).

A considerable number of cases of overt zinc deficiency has been reported in premature infants fed breast milk or receiving parenteral nutrition. It is thus evident that premature infants are at a relatively high risk of zinc deficiency. Reasons for this include (a) High fecal losses of zinc (b) Low body stores at birth (c) Relatively high requirements for tissue gain (Lothallar et al., 1985). Fetal growth retardation is not associated with low maternal plasma zinc and indeed a negative correlation between birth weight and maternal plasma zinc has been reported in humans (Hendrik, et al., 1984).

(b) KERATOGENESIS:

An association between chronic skin ulceration from different causes in man and subnormal plasma or serum zinc concentration has been observed in several studies. For example Halsted and Smith (1977) reported

mean serum zinc of 58+15 mcg/dl in patients with indolent ulcers.

Skin changes associated with zinc deficiency are well recognized in the hereditary condition of acrodermatitis enteropathica. Acquired zinc deficiency has been described in a breast fed preterm infant and when total parenteral nutrition is administered without zinc supplementation. Acquired zinc deficiency can result from reduced zinc absorption or increased losses due to gastro-intestinal surgery (Cope man et al., 1984) and (Bernard Henry, 1986).

(c) Wound healing:

Canker sores, tiny painful ulcers that can lock onto the inside of the mouth or the tip of the tongue for a week or more may be caused by zinc deficiency.

Zinc may heal another wound the wounded pride of a person branded by acne. Giving zinc to acne patients a Swedish researcher found that the mineral cleared up 85% of their pimples after twelve weeks.

But acne is not the only skin problem that zinc can treat - it can also cool down boils (Hambidge,1979). The enhancement of wound healing by zinc may stem from a heightened metabolic demand for this element for collagen synthesis in the process of tissue repair, with an increase in collagen synthesis and cross linking

explaining gains in wound tensile strength. But direct evidence for this is lacking (Anthony et al., 1977). Further more, zinc responsive difference in tissue repair could be related to differences in the rate of cell division and DNA production in rapidly regenerating tissues, since Prasad and Oberleas have shown that depressed activity of thymidine kinase, necessary for DNA synthesis and cell division, is an early metabolic defect of zinc deficiency.

d) Brain development and behavior:-

Zinc deficiency during the critical period for brain growth permanently affects brain function. When this deficiency is imposed throughout the latter third of pregnancy, brain size is decreased there is reduced total brain cell number, and the cytoplasmic nuclear ratio is increased implying an impairment of cell division in the brain during the critical period of maroneuronal proliferation (Sandstead et al., 1975). Lack of zinc in the body can wear away at an area of the brain, the hippocampus, causing confusion and dullness. Elderly people with a 'reduced coping capacity' may suffer from zinc deficiency (Bill Gottlieb, 1979).

e) Night Sight:

One common eye disorder that can be caused by zinc deficiency is night blindness, the inability of the eyes to adapt to darkness(John Feltman, 1979).

f) Atherosclerosis:

Indications have been obtained that zinc therapy can be beneficial in some cases of atherosclerosis. The mode of action of zinc in atherosclerosis is unknown. Hair and plasma zinc levels are usually subnormal in atherosclerosis and myocardial infarctions and the aortic wall has active turnover of zinc. This becomes even more active when the arterial wall is injured, in a manner similar to that seen in skin and muscle. Since atherosclerosis is thought to begin with some form of trauma it may that it is, in part, an expression of inadequate arterial repair(EJUnderwood, 1977)

g) Sickle cell disease:-

A number of studies have suggested that zinc deficiency occurs in sickle cell anemia specifically, the growth retardation, hypogonadism, abnormal dark adaptation and cell mediated immune disorders seen in sickle cell anemia patients have been related to a zinc deficiency, however, the mechanism involved has not been determined, lowered intake and malabsorption of zinc are possible causative factors and is known that sickle cell anemia patients excrete excessive zinc in their urine,

probably as a result of abnormal renal tubular reabsorption of zinc due to the sickling phenomena (Robinson et al., 1984).

h) Reproduction:

Mutch and Hurley (1978) showed that zinc deficiency imposed on rats during lactation, rapidly reduced plasma zinc levels and caused an impairment in milk production which was specifically due to the lack of zinc rather than to inanition. The zinc level in the milk was also reduced so that the pups received only half the amount of zinc and became zinc deficient, as evidenced by reduced plasma zinc levels, impaired growth and increased mortality. The zinc deficient females delivered that their litters with extreme difficulty, suffered excessive bleeding, and failed to consume after births or to prepare a nestsite. The effects of zinc deficiency in the female depend on the severity, timing and duration of the deficiency.

i) Carbohydrate and lipid metabolism:-

Zinc deficiency produces a prediabetic state, induces, the rate of insulin degradation at tissue level and might interfere with the insulin synthesis, storage and release. Zinc deficiency might be the initiator of physiological alteration in glucose metabolism resulting in a state of diabetes mellitus (Gupta, 1983). There is

degradation of the triglyeride reserves and a consequent increase in blood FFA concentration (Florence 1979).

j) Protein and nucliec acid metabolism:-

Zinc is involved primarily in nucleic acid and protein metabolism and hence in the fundamental processes of cell replication. Impaired DNA synthesis in the liver of zinc deficient rats has been demonstrated in several studies The total protein and RNA contents of the testis of zinc deficient rats are reduced and the testis of more severely zinc deficient rats contain lower concentrations of zinc RNA, DNA and protein and higher non_protein nitrogen levels and ribonuclease activity than either the restricted-fed or unrestricted-fed controls. The process of 'gene activation' requires zinc. Evidence has also been obtained of a continuing zinc requirement for DNA synthesis in cultured chick embryo and mammalian cells providing further evidence of a role for zinc in the regulation of cell multiplication (Rubin, 1977). Carboxy peptidase activity is found to be appreciably reduced in zinc deficient rats and to return rapidly to normal with zinc therapy (Kirchgenner, 1976).

k) Recently low zinc values of plasma, serum, hair, parotid, saliva and reduced activity of zinc metallo enzymes have been reported in several childhood disorders like Indian childhood cirrhosis, thalassemia, lung infections active tuberculosis, Downs syndrome, pica, and malnutrition (Srivatava et al., 1985)

Zinc deficiency also occurs in the treatment of hypercholesterolemia with ethylenediamine tetracetate. Penicillamine treatment of cystinuria, or of Willson's disease may have the same effect (Majumdar, 1984).

K) Host Zinc nutrition and Ehrlich Ascites tumor growth:-

Recent experiments with Zinc, are focussed on the mechanistic explanation of the inhibition of tumor growth by low zinc intake. Zinc deficiency is capable of inhibiting a variety of transplantable tumors, including the walker 256 carcinosarcoma, the levis lung sarcoma and leukemias. Although tumor growth could be limited by a host deficiency of zinc, established either before or upto 4 days after tumor cell inoculation, a similar deficiency established at a later time could not retard tumor growth. Thus the growth rate of an established colony of tumor cells is not altered by Zinc deficiency (Siddarth, 1984).

1) Zinc and environmental pollution:-

Studies have shown that Zinc can protect the body against the environmental pollutants lead and cadmium, speed the healing of stomach ulcers, improve the flexibility of rheumatoid arthritis and reduce a swollen prostate gland, a problem that afflicts almost all men over 60 (John Feltman, 1979).

4. REQUIREMENTS OF ZINC:

The minimum zinc requirement of human with satisfactory growth, health and well being vary with the type of diet

consumed, climatic conditions and the existence of stress imposed by trauma, parasitic infestations and infections. The National Academy of Sciences of United States of America (1974) has recommended the following daily dietary allowances of Zinc. Infants: 3-5 mg; Children 1-10 years; 10mg; males 10-51 + years 15 mg. females 10-51+ years 15mg. pregnant women 20mg; and lactating women 25 mg.

5. ZINC TOXICITY:

The relatively low toxicity of zinc among the divalent cations, coupled with efficient homeostatics control mechanisms, make chronic Zinc toxicity from dietary sources an unlikely hazard to man. Where zinc salts or compounds are given orally in large doses over prolonged periods, as in the treatment of chronic leg ulcers or the prophylaxis of cardiovascular disease, possibilities of toxic effects cannot be dismissed (Lanner et al., 1977). Zinc is relatively non toxic to birds and mammals. Rats pigs, poultry, sheep, cattle and men exhibit considerable tollerance to high zinc intakes, the extent of the tolerance depending greatly on the nature of the diet particularly its content of calcium, copper, iron and cadmium, with which it interacts in the process of absorption and utilization (Lanner et al., 1977).

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B. SELENIUM:

Selenium is in all probability required by the human organism, just as it is for all experimental animals studied. It certainly appears to be an important nutrient for man like the better known trace elements such as Zinc, copper and manganese (Jean Bogert, 1977).

1. SOURCES OF SELENIUM:

(a) Human foods and dietaries: The level of selenium in individual foods of plant orgin is highly variable depending mainly on the soil conditions under which they are grown (Hoekstra et al., 1975).

Seafoods, kidney, liver, meat and whole grains are generally good sources (0.2 ppm selenium) and fruits and vegetables are mostly poor sources (0.01 ppm selenium or less wet weight) (Levander et al., 1980). Fish flour for human consumption has been reported to contain 1.8 ppm selenium and tunafish meal median levels (dry basis) as high as 5.1 and 6.2 ppm (Thompson et al., 1975). Selenium is lost in the refining of sugar. Morris and Levander (1980) report a level of 0.012 for brown sugar and 0.003 mcg selenium/g for white sugar. Water supplies do not normally constitute a significant source of selenium to man, either in selenium deficient; 'normal' or 'seleni ferous areas', although levels in water from some seleni-

ferous areas as high as 50-300 mcg/litre have been reported (W.H.O., 1973. Mushrooms are relatively high in selenium (Mark Bricklin, 1979).

b) Animal feeds and forages:

The value of the different feeds stuffs as a source of selenium depends on the form in which the element is present, as well as on its concentration, at least for chicks and rats. Plant sources generally have a much higher selenium availability than animal products (Noguchi et al.,1975) Much of the selenium in wheat and probably other grains and in alfalfa is in the form of selenomethionine (Plamer et al., 1979). Little of the selenium in animal products is likely to be in this form, although it is largely associated with the protein fraction. The mean selenium concentration of all forages was 0.19ppm with no significant differences between grasses and clovers and of all grains 0.27 ppm with wheat significantly higher (0.32 ppm) than barley and oats (0.2ppm) Much lower selenium levels in cereal grains from deficient areas, down to 0.006-0.007 ppm have been reported (Thompson et al.,1977).

2. OCCURRENCE OF SELENIUM IN TISSUES AND FLUIDS:

(i) General distribution:

The liver and kidney usually carry the highest selenium concentrations, with much lower levels in the muscles, bones and blood and very low levels in adipose tissue. Cardiac muscle is consistently higher in Selenium than skeletal muscle (Godwin et al., 1977). Selenium concen-

trations in the tissues reflect the level of dietary selenium over a wide range comparatively few data have appeared on the Selenium levels in normal human tissues other than blood. Dickson and Tomlinson (1978) reported the following values for autopsy specimens for adults: liver 0.18- 0·66 skin 0.12-0.62 and muscle 0.26-0.59 mcg selenium per g whole tissue. In a later investigation carried out in England (1977) the mean selenium concentration in mcg/g whole tissue were reported as follows:

Liver 0.30± 0.10: Kidney 0.10 ± 0.20: muscle, 0.11 ± 0.01 lung 0.10± 0.20; brain, 0.09± 0.02 testis 0.20± 0.04 and ovary 0.09±0.03. The selenium level in human dental enamel ranged from 0.12 to 0.90 mcg/g dry weight (Brown, et al.,1975). The high selenium content of human finger nails compared with other normal tissues has been pointed out by Hadjimarkos and Shearer (1975). By contrast these workers found the selenium concentration of the saliva of a group of children to be very low (range 1.1 - 5.2 ppm).

(ii) Selenium in blood:

The concentration of selenium in blood is highly responsive to changes in the selenium level in the diet over a wide range. The selenium levels in whole human blood ranges from 0.10 to 0.34 mcg/ml (Roth et al.,1983) McConnel and Coworkers (1975) reported a study of selenium in human blood sera in health and disease. Their mean

3. THE BIOCHEMISTRY OF SELENIUM:

(i) Absorption:

Studies with ^{75}Se at physiological levels indicate that the doudenum is the main site of selenium absorption from the rumen of abomasum of sheep or the stomach of pigs (Bell et al., 1977) in a study of the long term fate of an oral dose ^{75}Se selinite in three young women it was found that intestinal absorption was 70,64 and 44% of the dose (Stewart et al., 1974). Absorbed selenium is at first carried mainly in the plasma from which it enters all tissues, including the bones hair and leukocytes (Berry et al., 1977). Selinite Selenium has to undergo a chemical transformation by the erythrocytes in order to be bound by the plasma proteins. The process of expulsion of Selenium from the erythrocytes depends on adequate glutathione levels in these cells. Most of this selenium was initially observed to be transported by albumin after which it moved to the globular fractions. Sandholm (1976) has shown that the selenium that has been processed by the erythrocytes is taken up largely by β-lipoprotein and an unidentified fraction located electrophoretically between α_1 and α_2 globulin fractions.

(ii) EXCRETION:

Fecal excretion of ingested selenium is generally greater than urinary excretion in ruminants but not in monogastric species. Most of the selenium in the feces

consists of unabsorbed dietary selenium together with small amounts excreted into the bowel in the biliary, pancreatic and intestinal secretions. Lavender and Vaumann(1977) found that selenium excretion into the gastro intestinal tract via the bile fluid is markedly increased and retention in the carcass, liver and blood greatly decreased with no effect on urinary excretion when subacute. Injections of Mercury, thallium and lead had no such effect ~~on effect~~ on biliary selenium excretion (Higgs et al.,1974) Arsenic injections given prior to the selenium reduce urinary selenium excretion in rats and a new selenium metabolite identified as trimethylsenelonium ion has been isolated from the urine of rats injected with (^{75}Se) selenite (Olson et al., 1977).

(iii) Functions and effects of selenium deficiency:

Selenium has been shown to be an essential trace element, necessary for enzymatic activity of glutathione peroxidase (GSHPX). GSHPX catalyses the degradation of hydrogen peroxide and organic hydroperoxides. In the absence of selenium GSHPX is not active and lipid peroxides and free radicals may damage cell membranes (Marilyn R.Brown et al., 1986). Glutathione peroxidase (GSHPX) was identified as a selenoprotein by Rotruck et al., in 1983 and in the same year Flohe and co-workers showed that the selenium in the enzyme was in stoichiometric amounts with 4 g atoms Se/mole.

$$2\ GSH + H_2O_2 \xrightarrow{\text{Glutathione peroxidase}} GSSG + H_2O$$

$$GSSG + NADPH \xrightarrow{\text{Glutathione reductase}} 2\ GSH + NADP^+$$

Glutathione peroxidase activity has been demonstrated in a wide range of body tissues, fluids, cells and sub-cellular fractions at levels which vary greatly with the species, tissue, and selenium status of the animal. The highest GSHPX activity commonly occurs in the liver moderately high activity in the erythrocytes, heart muscle, lung, and kindneys and smaller activity in the intestinal tract and skeletal muscle (Marilyn R.Brown et al.,1986). Although the need for degradation of H_2O_2 and organic hydroperoxides by glutathione peroxidase is important, the absence of the activity of this enzyme in selenium deficient patients does not automatically indicate the danger of developing any particular manifestation possibly because Vitamin-E can prevent peroxidation of tissues at the membrane level. The combination of selenium and Vitamin-E deficiency may be required in some patients for the symptoms to develop. The serum Vitamin-E level was within normal limits (William Jklish et al., 1986).

Symptoms related to selenium deficiency reported in humans include muscle pain and cardiomyopathy. Lower extremity thigh muscle pain was described in one patient by Vanrij et al., 1979. The patient muscle pain responded to selenomethionine given intravenously.

3. Exudative diathesis:

Chicks fed selenium deficiency diets fail to grow and develop a diseased condition known as Exudative diathesis.

4. Pancreatic fibrosis:

Severe selenium deficiency has been shown by Scott and his collaborators to result in atrophy of the pancreas of chicks, in addition to poor growth and feathering even in the presence of dietary Vitamin-E.

5. Hepatosis Dietetica in pigs:

Hepatosis dietetica has been produced on vitamin-E free diets based on Torula Yeast or soybean meal and occurs spontaneously in Newzealand and Scandinavia when pigs are fed grain rations naturally selenium low. The disease is most common at 3-15 weeks of age and results in high mortality. Severe necrotic liver lesions are apparent at postmortem examination. There is also deposition of steroid pigment in adipose tissue, giving a yellowish brown color to the body fat and a generalised subcutaneous edema (Merilyn et al., 1986).

6. Selenium Responsive Unthriftiness in Sheep and Cattle:

In parts of Newzeland a serious condition known as "ill thrift" occurs in lambs at pasture and can occur in beef and dairy cattle of all ages. The condition varies from a subclinical growth deficit to clinical unthriftness with rapid loss of weight and sometimes mortality. Ill thrift can be prevented by selenium treatement with striking increases in growth (Grant et al., 1979).

7. Immunological Responses:

Dietary selenium at levels above those generally accepted as adequate (0.1ppm) enhance the primary immune response in mice, as measured by the PFC (Plaque forming cell) test and by hemagglutination (Heinzerling et al., 1985). Sodium selenite administered to mice intraperitoneally at about 5 mcg selenium was shown to enhance the primary immune response to the sheep red cell blood antigen, the greatest enhancement occuring when selenium was administered with or prior to the antigen (Gerlach et al., 1979).

Selenium and Vitamin-E are both essential for curing certain diseased states in experimental animals such as (1) Necrosis of liver in rats and pigs (2) Exudative diathesis in birds and (3) Muscular dystrophy in cattle.

9. Selenium and dental caries:

In the oregon study children born and reared in a country where selenium deficiency in livestock is common, had a lower incidence of caries than children from other countries of known higher selenium status. It seems that incoporation of dietary selenium into the protein fraction of the enamel during the development of teeth may inhibit mineralization of tissue resulting in an increased susceptibility to caries (Hadjimarkos et al.,1973). Inhibition of growth and calcification of the teeth and maxillary bone by selenium injections as selenite has also been demonstrated in young rats, when the selenium doses exceed (Petrovic et al., 1977).

4. REQUIREMENTS OF SELENEIUM:

The selenium requirements of man are unknown. No pathological conditions unequivocally resulting from selenium deficiency have been identified in human individuals even in areas where this deficiency is severe and widespread in livestock. In the subhuman primate (saimiriscioureus) selenium deficiency is characterized by loss of weight, listlessness and alopecia has been produced by feeding a low selenium Torula yeast diet adequate in Vitamin-E for nine months (Oldfield et al., 1979). Non treated monkeys died and showed cardiac and skeletal muscle degeneration and hepatic necrosis. Monkeys treated with small (0.04 mg selenium sodiumselenite) injection at

two week intervals recovered rapidly. The minimum selenium requirements of animals vary with the form of the selenium ingested and the nature of the rest of diet (Hodgson et al., 1980).

SELENIUM AND CANCER:

There are several epidemiological studies suggesting an increase in incidence of colon, mammary and perhaps other forms of cancer associated with low levels of environmental selenium. Selenium compounds added to the diet or water have now been shown to be effective inhibitors of chemical carcinogenesis in different experimental animals. (Schnauzer and Ishmael, 1977). Some scientists suggest that selenium may possess cancer protecting properties in the humans (Leklem et al., 1983). Human female breast cancer mortalities have been inversely correlated with selenium concentration in whole blood (Schneider et al., 1977). Spallholtz et al.,(1977) noted enhanced immune response in selenium treated rats and mice. This effect may be associated with tumorigenesis prevention in the early phases of cancer development. "Optimal cancer protection" would be provided approximately by 300 mcg selenium/day/person and with less than 150 mcg/day having no or minimal protecting effect. A human diet rich in seafoods and cereals may lower the risk of cancer when compared to predominantly meat based diet (Shultz et al., 1983). Clayton and Baumann (1978) found that

the inclusion of 5 ppm selenium as selenite in a purified diet reduced the incidence of liver tumors in rats which had previously received a carcinigenic azo dye. Injection of 1 mg of selenocystine/kg body weight for 14 days following injection of Murphy's lymphosarcoma was later shown to significantly reduce the average size of the tumors in rate and the feeding of selenium inhibited the carcinogenic effect of croton oil in Mice (Rudolph et al., 1977). Human cancer incidence and mortality could be lowered by appropriate dietary selenium supplementation in low selenium areas.

B. SELENIUM TOXICITY:

Selenium in excess is very toxic. In some areas of the Western united states and Newzealand excess selenium in the soil and consequently in the vegetation is the cause of "blind staggers" in horses and "alkali disease" in cattle (Lehninger, 1984).

a) Manifestations of selenenosis:

All degrees of selenium poisoning exist, from a mild chronic condition to an acute form resulting in death of the animal. Chronic selenium poisoning is characterised by dullness and lack of vitality, emaciation and roughness of coat, loss of hair from the mane and tail of horses and the body of pigs, soreness and sloughing of the hoofs, stiffness and lameness due to erosion of the joints of the long bones, atrophy of the heart ("dish-rag" heart), cirrhosis of the liver and anemia. In acute sele-

nium poisoning the animals suffer from blindness, abdominal pain, salivation, grating of teeth and some degree of paralysis. Respiration is distrubed and death results from respiration failure (Jones et al., 1977). Nails become brittle and white spots and longitudinal streaks appear in the surface followed by a break on the wall of the nails. Thumbs are always affected first. As new growth continues the broken nails is pushed forward and finally drops off. This process may require one month to complete. In many cases fluid effuses from the skin around the nail. In those cases, a much longer period is needed to finish the whole process rough and stripped. Repeated attacks may result in acrophahia (clubbing of the fingers) (Shuzhuangsum et al., 1983). Skin lesions occur mainly on the four limbs ie., the back of hands and feet the outerside of legs and thighs, the forearms and the back of the neck. Affected skin becomes red and swollen and then blistered and eruptive (cuevas et al., 1972).

Abnormalities of the nervous system were observed only in heavily affected village with selenosis. Initially patients may complain of peripheral anesthesia "Pins and needles" acroparaesthesia and pain in the extremities. Hyperreflexia of the tendon is common, and then numbness, convulsions paralysis, motor disturbance and even hemiplegia may develop. It is likely that all clinical neu-

rological signs are due to polyneuritis caused by intoxication. Disturbances of the digestive tract usually accompanied this type of intoxication (Shuzhuangsun et al., 1983).

b. Factors affecting Selenium toxicity:

The toxicity of selenium to animals varies with the amounts and chemical forms of the selenium ingested, the duration and continuity of the selenium intake, the nature of the rest of the diet and to some extent with the species (Bowen, 1972). The toxicity of selenium can be greately modified by the dietary levels of arsenic, silver, mercury, copper and cadmium with each element apparently copper and cadmium ~~with each element apparently~~ exerting its protecting action by its own mechanism. The effect of arsenic was first demonstrated by Moxon who showed that 5 ppm arsenic as arsenite in the drinking water prevented all signs of selenosis in rats.

c. The mechanism of selenium toxicity:

The precise ways in which selenium at toxic intakes interferes with tissue structure and functions are not completely understood, Selenium has long been known to affect certain unicellular organism and enzyme systems to inhibit alcoholic fermentation by yeast and some of the enzymes concerned with cellular respiration(Elvehjem et al., 1977). The inhibition of oxygen consumption by

tissues appears ~~to be~~ tissues ~~appears~~ to be mediated through a poisoning of succinic dehydrogenase. The liver succinic dehydrogenase levels of rats fed seleniferous diets are reduced below normal and can be maintained at normal levels by appropriate dietary intakes of arsenic. It is unlikely that these effects are sufficient to account for the various manifestations of selenosis or their prevention by arsenic (Potter et al., 1977)

C. CHROMIUM:

1. CHROMIUM SOURCES AND REQUIREMENTS:

Table 2 presents the Chromium Content of foods.

TABLE 2

CHROMIUM CONTENT OF FOODS (JOSEPH HUBERT et al., 1979)

	Food Type	Chromium content ppm
1.	Milk, Dairy products	0.05
2.	Meat, fish poultry	0.06
3.	Cereals	0.06
4.	Potatoes	0.04
5.	Leafy vegetables	0.09
6.	Legumes	0.06
7.	Root Vegetables	0.09
8.	Garden fruits	0.05
9.	Fruits	0.05
10.	Oils and fats	0.03
11.	Sugars	0.34
12.	Drinks	0.03

It appears from Table-2 that most food stuffs contain Chromium, but with the exception of yeast none is a particularly rich source. The minimum human chromium requirements compatible with satisfactory growth and long term health and fertility cannot yet be given because of inadequate knowledge of the forms and availability of chromium in foods. The chromium content of diets of many healthy adults has recently reported as subsequently less than 50mcg/day (F'xavier Pi-Sunyer, 1986). This is below the recommended daily intake of 50 to 200 mcg that was proposed as safe and adequate in 1980 by the Food and Nutrition Board of the National Academy of Sciences (Friden et al., 1984). If healthy adults indeed consume less than 50 mcg chromium/day, it is important to ascertain whether they represent a risk of chromium dificiency. It is very difficult to determine precisely how much chromium the body needs to maintain a normal balance because the very low levels in blood and excreta pose severe analytical problems (Anderson et al., 1984).

2. OCCURRENCE OF CHROMIUM IN TISSUES AND FLUIDS:

Chromium is widely distributed throughout the human body in low concentrations without special concentration in any known tissue or organ and these levels decline with age, except in lungs. The levels decline rapidly in the first decade of life in the heart, lung, aorta and

spleen, while in the liver and kidney the neonatal concentrations are maintained until the second decade, when a decline occurs. Substantial variations in human liver and kidney chromium levels have been observed in different geographical regions, presumably as a reflection of regional differences in environmental chromium intakes (Clayton et al., 1984).

The hair chromium levels are highest at birth, decline rapidly in childhood and less so in the adult. The hair chromium levels do not increase with the length of hair showing little environmental contamination. The diabetic subjects have lower hair chromium levels. The pregnant women have lower hair chromium levels than non-pregnant women. Hair chromium level is higher at one year of age than subsequently, although the values vary considerably with age within the individual (Joseph Hurbert, 1979).

Mean levels of 57 and 13ng chromium/g were reported for cow's colostrum and normal milk, respectively. (Hambidge (1979) examined 14 samples of breast milk from five women and reported a mean of 11.6 (Range 6.4-18.5) ng chromium/ml.

Very few data on the chromium content of eggs are available. Kirkpatrick and Coffin (1975) analyzed 100 Shilled chicken egg samples. The chromium concentrations reported for these samples ranged from less then 0.05 to

o.15 with a calculated mean of 0.06 mcg/g fresh basis Much effort has been directed towards the determination of chromium in serum. However widely divergent values have been reported, varying from less than 0.50 ng/ml to 41-251 ng/ml. Previously reported plasma or serum concentrations in healthy individuals are as follows:-

T A B L E IV

Reference	Year	Analytical Technique	Normal value ng/ml
Seeling et al.,	1975	AAS	0.73 (0.23-1.90)
Grafflage et al.,	1974	AAS	0.73 (0.23-1.90)
Pekarek et al.,	1974	AAS	1.62
Liu and Morris	1978	NAA	1.67 (-)
Hambidge,	1974	ESM	3.1
Davidson and Secrest,	1972	AAS	5.07 (3.10-7.19)
Freund et al.,	1979	Unknown	5-90
Li and Hecules	1974	CHL	150 (41 - 251)

Abbreviations used : AAS - Atomic Absorption Spectrometry
CHL - Chemiluminescence
ESM - Emission spectrometry

J.H.Dennis Shapcott et al., (1979) found a mean serum chromium concentration of 0.160ng/ml. According to Dennis Shapcott et al., (1979) Serum Chromium concentration in normal subjects must be lower than 0.5 ng/ml and Inconsistencies between the results of different authors are mainly due to contamination.

3. THE BIOCHEMISTRY OF CHROMIUM:

(i) Absorption:

It has been reported that less than 2% of the Chromium contained in the diet is absorbed from the intestine (Hillary, 1986). Inorganic chromium compounds are poorly absorbed in animals and man, to the extent of 1-3% or less, regardless of the dose and dietary chromium status. Chromium is absorbed in the small intestine by a pathway it appears to share with zinc. It is transported to tissues bound to transferrin and appears in liver mitochondria, microsomes and the cytosol. Hexavalent chromium is much more toxic than the trivalent chromium, Chronic occupational exposure to chromate dust seems to carry an increased risk of the lung cancer. Significant chromium is contributed to the diet by cooking in stainless steel cookware (Dryl K.Granner et al., 1985).

(ii) Excretion:

Chromium is excreted mainly in the urine, whether ingested or injected although small amounts are lost in the feces via the bile and small intestine and possible through the skin. The total amount of chromium normally excreted in urine averages less than 0.5 mcg/day. Further more the variability of urinary chromium from day to day in free living people seems to be affected by customary daily variations in dietary intake and living habits and by the health status(Clayton et al.,1984).

(iii) Functions and effects of chromium deficiency:

Chromium is an essential trace element for humans and animals and is required for normal Carbohydrate, lipid and protein metabolism. Insufficient dietary chromium leads to signs and symptoms similar to those observed for diabetes and cardiovascular diseases.

(a) Carbohydrate metabolism:

Chromium is thought to play some functional role in the regulation of glucose metabolism. The trivalent form of chromium (cr^{3+}) can improve the glucose tolerance of individuals suffering from Protein calorie malnutrition (Grenner et al., 1985). Fractionation studies of brewer's yeast yielded fractions with much greater biological activity then chromium as $CrCl_3$, suggesting the existence of a chromium containing complex Glucose Tolerance Factor (GTF) (Metrz et al., 1979).

Severe chromium deficiency in human subjects exhibiting weight loss, peripheral neuropathy impaired Glucose tolerance and subnormal blood and hair chromium concentrations, while on prolonged total parenteral nutrition, and responsive to chromium therapy has been reported (Brusce Robertson et al., 1975). The results indicated that isolated chromium deficiency in man causes glucose intolerance, inability to utilise glucose for

energy neuropathy with normal insulin levels and no impairment of insulin action on amino acid uptake and FFA release. It is apparent that many individuals are ingesting insufficient chromium to maintain normal glucose utilization. The fact that not every malnourished infant or every diabetic or old person responds to chromium supplementation indicates that chromium status of the nonresponders is adequate for this function or that one of the many other factors influencing carbohydrate metabolism is operating in these cases (E. Underwood, 1977).

(b) Lipid metabolism:

There is some evidence that chromium plays a role in serum cholesterol homeostasis. The addition of chromium to a low chromium diet suppressed serum cholestrol levels in rats and in males inhibited the tendency of these levels to increase with age. Further, a depression in serum cholestrol was achieved in male rats by feeding 1 mcg/chromium/ml in this way (Schroeder, 1977).

(c) Protein Synthesis:

Rats fed diets deficient in Chromium and protein have an impaired capacity to incorporate several aminoacids into the protein of their hearts (Mertz et al., 1979). Slightly improved incorporation was achieved with insulin alone which was significantly enhanced by chromium(iii) supplementation. The amino acids affected by chromium

were α amino isobutyric acid, glycine, serine and methionine. No such effect of chromium was observed with lysine, phenylalanine, and a mixture of ten other amino acids.

Insulin invivo also stimulated the cell transport of an amino acid analog to a greater degree in rats fed a low protein chromium supplemented diet than it did in chromium deficient controls (Roginski et al., 1977). The claim that chromium acts as cofactor for insulin can therefore also be applied to 2 insulin responsive steps in amino acid metabolism which are independent of the action of insulin on glucose utilization.

4. Growth and Longevity:

On a diet of rye skim milk and corn oil with added vitamins and Zn, Cu, Mn, Co and MO in the water, male mice and rats receiving 2 or 5 ppm (Chromium III) in the drinking water grew significantly better than controls. The median age of male mice at death was 99 days longer when they were fed chromium than when they were not and the mean age was 91 days longer. No such differences in longevity due to chromium were observed in female mice or rats. The chromium treatment had no effect on the incidences of tumors but appeared to protect female rats against lung infection (Balassa et al., 1979).

(4) CHROMIUM TOXICITY:

Hexavalent chromium is much more toxic than trivalent. In fact trivalent chromium has such a low order of toxicity that a wide margin of safety exists between the amounts ordinarily ingested and those likely to induce deleterious effects. Lifetime exposure to 5mg/litre of chromium (III) in the drinking water induced no toxic effects in rats and mice and exposure of mice of three generations to chromium oxide at levels upto 20ppm of the diet had no measurable effect on mortality, morbidity, growth or fertility (Luckey et al., 1978). Chronic exposure to chromate dust has been correlated with increased incidence of lung cancer, and oral administration of 50 ppm of chromate has been associated with growth depression and liver and kidney damage in experimental animals. Toxic exposure to the skin results in dermatitis and persistent ulceration. Accidental ingestion has resulted invertigo abdominal pain, vomiting, anuria, convulsions, shock and coma (John et al., 1986).

* * * * * *

Experimental Procedure

III. EXPERIMENTAL PROCEDURE

The experimental procedure pertaining to the study 'Serum Zinc, selenium and chromium levels of normal persons' is discussed under the following headings:

1. Selection and grouping of normal persons
2. Collection of blood samples
3. Separation of serum
4. Estimation of serum zinc level
5. Estimation of serum selenium level
6. Estimation of serum chromium level
7. Statistical analysis.

1. Selection and grouping of normal persons:

A group of one hundred and sixteen normal healthy persons of both sexes consisting of doctors and nurses of E.S.I. Hospital, Singanallur and the donors of Rayvijay Blood Bank and E.S.I.Hospital blood bank were selected for the study. Table I presents their distribution according to age and food pattern.

T A B L E - I

Grouping of Normal Persons

	Age in years			
	15-20	21-40	41-60	Total
	No. of persons			
Vegetarian				
Males	10	11	9	30
Females	11	9	8	28
Non-vegetarian				
Males	7	13	7	27
Females	9	12	10	31
Total	37	45	34	116

2. Collection of blood samples:

The blood was collected as follows (Oser 1976): Tied a tourniquet (of soft rubber tubing or strip of bandage tightly around the arm of the persons, a couple of inches above the elbow. Had the subject clench his fist firmly, washed the skin surface above the promient vein on the inner surface of the elblow (usually median bassillie) with 70% alcohol, allowed to dry, held the vein immobile by pressing on it with the thumb below the elbow and into the vein, inserted a sharp sterile hypodermic needle (No:22) an inch or a half long which was attached to dry sterile syringe of suitable capacity.

The needle should penetrate the vein from the side and at an angle of 50° with the surface of the arm, the level of opening of the needle being kept upwards or to the side. As soon as blood was seen to enter the syringe, retracted the plunger slowly until the desired amount of blood had entered the syringe. Before removing the needle from the vein, loosened the tourniquet, had the patient unclench his fist and on the skin, at the point of entrance of the needle held inplace a small pad of folded gauze moistened with 70% alchohol. Withdraw the needle, detached it from the syringe (not too vigorously which might cause hemolysis) and then transferred the blood to a centrifuge tube.

3. Separation of Serum:

Blood was transferred to clean empty centrifuge tubes and allowed to clot for three hours at room temperature. The clot was allowed to retract. Then it was centrifuged and the serum was separated using a rubber bulb pipette. It was collected in clean dry labelled test tubes and stored in the freezer until used.

4. Estimation of serum Zinc level:

Serum zinc level was estimated by atomic absorption spectrophotometer (Piper's method, 1966)

Principle:

Serum on digestion with triple acid, Nitric acid, sulphuric acid, and perchloric acid in the ratio of 9:2:1 liberates into solution the trace elements.

Procedure:

1.0 ml of the serum sample was taken in a microk-jeldahl digestion flask which was previously washed with glass distilled water and dried, and to this was added 10.0 ml. of triple acid. The mixture was shaken and digested on a sand bath with occasional shaking the digestion was continued till no more brown fumes evolved and the solution in the flask became colourless. The digested mixture was transferred to a 25.0ml standard flask, the washing being done with double distilled water. The solution was used for analysing serum zinc level using the atomic absorption spectro-photometer (AARO model) available in the soil science Department in Tamil Nadu Agricultural University, Coimbatore.

5. Estimation of Serum Selenium:

Selenium was estimated by colorimetric method of Snell, D. and Snell T. (1967)

Principle:

Selenium in an acid solution such as hydrochloric acid is reduced to the element with a suitable reagent like sodium metabisulphite and the resulting orange to red colour is read colorimetrically.

Reagents:

1. Selenium powder
2. Concentrated nitric acid.
3. Concentrated hydrochloric acid.
4. Saturated sodium metabisulfite solution.

All the reagents were prepared in doubled distilled water.

Standard Selenium Solution:

100 mg of selenium powder was weighed and a few drops of concentrated nitric acid was added and then evaporated to dryness. This method was repeated twice. Dissolved the dried salt in 10.0 ml of dilute hydrochloric acid (1:9) and made up to 100 ml with the same.(AOAC,1980)

Working Standared:

1.0ml of selenium stock solution was made upto 1000ml with distilled water so that 1.0ml of this solution contained 1.0 µg of Selenium. Again 10.0ml of this solution was diluted to 100ml so that 1.0ml of the solution contains 0.1 µg.

Serum Treatment:

To 1.0ml of serum added a few drops of concentrated nitric acid and evaporated to dryness. 9.0ml of water was added and then evaporated to dryness. The procedure was repeated twice and then made upto 25.0ml with dilute hydrochloric acid(1.0ml hydrochloric acid and 9.0ml water)

Procedure:

0.2, 0.4, 0.6, 0.8 and 1.0ml of selenium standard solution was pipetted out into clean dry test tubes. 1.0ml of glycerol was added to each tube to stabilise the solution. Then to each of the above tubes saturated metabisulphite solution equivalent to double the volume of selenium standard solution was added. For instance to the first tube which contained 0.2ml of selenium standard solution 1.0ml of metabisulphite was added. Heated the tubes for seven minutes at 70°C Cooled and made upto 5.0ml in all the tubes with water and read the tranmittance at 420 nm. 1.0ml of the treated serum was taken and repeated the above procedure.

6. Estimation of Serum Chromium:

Chromium was estimated by colorimetric method by James W.M.C. Coy (1969). The details of the method is given below:

Principle:

Chromium is oxidised by permanganate to chromate and then treated with diphenyl carbazide to obtain the violet coloured complex which is measured colorimetrically.

Inferences:

Mercury and vanadium interfere but they occur in negligible concentrations, Iron interferes when its concentration exceeds 1 mg/litre but can be estimated by

preliminary treatement with cupferon.

Reagents:

Double distiled water was used for the preparation of reagents.

1. Chromium stock solution:

Dissolved 283mg dried potassium dichromate ($K_2Cr_2O_7$) in distilled water and made upto 100ml in a volumetric flask. 1.0 ml contains 100 µg of chromium.

2. Chromium working standard:

10.0ml of chromium stock solution was diluted to 1000ml in a standard flask so that 1.0ml of the made up solution contains 1.0 µg of chromium. 1.0ml of this was again diluted to 1000ml so that 1.0ml of the solution contains 0.001 µg of chromium.

3. Phosphoric acid 85%

4. Sulphuric acid 5%:

Carefully added 50.0ml of concentrated sulphuric acid to 950ml distilled water.

5. Diphenyl carbazide solution:

Dissolved 500 mg diphenyl carbazide and 8.0g of Phlthalic anhydride in 200 ml 95% ethylalcohol. Placed it in a amber bottle and kept it in refrigeration. (Reagent is stable for several months in a refrigerator).

6. Potassium permanganate:

Appriximately 0.1N. Dissolved 316 mg potassium permanganate in little of distilled water and diluted to 100ml.

7. Sodium azide solution:

Dissolved 500mg sodium azide ($Na\ N_3$) in 100ml chromium free distilled water.

Reagents for destruction of organic matter:-

1. Sodium azide 2. Concentrated Sulphuric acid 3. Concentrated Nitric acid 4. Saturated ammonium Oxalate solution 5. Concentrated ammonia solution.

Procedure:

Serum was completely oxidised by low temperature ashing and it gave a residue that was completely soluble in dilute acid.

Destruction of organic matter in the sample:

To destroy the organic matter suitable volume of the sample containing not more than 50 µg is placed in a 250ml Kjeldhal flask. Dissolved 100 mg sodium sulphite in it and added 2.0 ml concentrated sulphuric acid. Evaporated until white fumes evolved. If the organic matter is hard to destroy added 1.0 to 2.0ml of concentrated nitric acid in dropwise. Added 10.0ml of saturated ammonium oxalate solution and evaporated to

fuming again until a minimum residual acid liquor is obtained. Cooled and diluted with 10ml of distilled water and transferred the contents in the Kjeldahl flask to a 25.0ml volumetric flask and made upto the mark with distilled water. Placed the whole of the made up solution or an aliquot in a 50ml beaker. Neutralised with concentrated ammonia solution.

To the above along with the standard added 0.5ml of phosphoric acid, 10.0ml of 5% sulphuric acid and 4.0ml of diphenyl carbazide. The pink color developed was read at 540nm.

7. Statistical Analysis:

Students 't' tests were conducted wherever necessary to check it the results were significant using the formula:

$$t = \frac{\bar{X}_1 - \bar{X}_2}{\sqrt{\frac{(n_1 s_1^2) + (n_2 s_2^2)}{n_1 + n_2 - 2} \left(\frac{1}{n_1} + \frac{1}{n_2} \right)}}$$

$\bar{X}_1$ and $\bar{X}_2$ = mean of 1st and 2nd sample

s_1 and s_2 = Standard deviation of n_1 and n_2 sample

n_1 and n_2 = Number of observation of the 1st and 2nd samples.

Results and Discussion

IV. RESULTS AND DISCUSSION

The results and discussion pertaining to the study "Serum zinc, selenium and chromium levels in normal persons" are discussed under the following headings:-

1. Distribution of normal persons according to age, sex, and pattern of diet.

2. Levels of zinc in serum:
   a) Effect of age
   b) Effect of diet
   c) Effect of sex

3. Levels of selenium in serum:
   a) Effect of age
   b) Effect of diet
   c) Effect of sex

4. Levels of chromium in serum:
   a) Effect of age
   b) Effect of diet
   c) Effect of sex

****

1. THE DISTRIBUTION OF NORMAL PERSONS ACCORDING TO AGE, SEX AND PATTERN OF DIET:

Table - 1 presents the distribution of normal person according to age, sex, and pattern of diet.

T A B L E - 1

DISTRIBUTION OF NORMAL PERSONS ACCORDING TO AGE, SEX AND PATTERN OF DIET.

Group	Age in Years			Total
	15-20	21-40	41-60	
	Number of persons			
Vegetarians				
Males	10	11	9	30
Females	11	9	8	28
Non-vegetarians				
Males	7	13	7	27
Females	9	12	10	31
Total	37	45	34	116

2. Levels of zinc in Serum

(a) Effect of age:

Table -2 presents the comparison of serum zinc levels among different age groups of the same sex and same pattern of diet.

TABLE - 2

Serum zinc levels in different age groups in/µg/dl

Diet pattern	Sex	Age	Group	Mean±S.D	Group compared	't' Value
I. Vegetarian	M	15-20	(a_1)	95.3±4.77	a_1 Vs b_1	1.053N.S
	M	21-40	(b_1)	97.54±4.5	a_1 Vs c_1	0.343N.S
	M	41-60	(c_1)	94.6±3.46	b_1 Vs c_1	1.531N.S
	F	15-20	(x_1)	95.9±2.71	x_1 Vs y_1	1.142N.S
	F	21-40	(y_1)	97.2±1.98	x_1 Vs z_1	0.749N.S
	F	41-60	(z_1)	94.9±2.962	y_1 Vs z_1	1.785N.S
II. Nonvegetarians:						
	M	15-20	(a_2)	114.1±16.7	a_2 Vs b_2	1.76N.S
	M	21-40	(b_2)	124.5± 18.8	a_2 Vs c_2	1.178N.S
	M	41-60	(c_2)	112.4±16.25	b_2 Vs c_2	1.370N.S
	F	15-20	(x_2)	106.0± 10.5	x_2 Vs y_2	3.146**
	F	21-40	(y_2)	129.6±19.4	x_2 Vs z_2	0.287N.S
	F	41-60	(z_2)	108.0±17.08	y_2 Vs z_2	2.673*

** Significant at 1% level

* Significant at 5%

N.S. Not significant.

From Table - 2, it is clear that the difference in the mean serum zinc values is not statistically significant among different age groups of men of both vegetarian and non-vegetarian patterns of diet. So age has no effect on serum zinc level of men consuming similar diet. However, among non-vegetarian women of the age group 21-40 the serum zinc level rises significantly at 1 percent level when compared with 15-20 age group the zinc level then falls in the age group 41-60, the fall being significant at 5% level. Thus the non-vegetarian female group of 21-40 years has the highest serum zinc level. While among vegetarian women age has no influence on serum zinc level as there is no satistically significant change among the different age groups.

(b) Effect of diet:

Table - 3 presents the comparison of serum zinc values between vegetarians and non-vegetarians of the same age group and sex.

T A B L E - 3

Serum zinc Levels of normal persons consisting of different patterns of Diet in |µg|dl.

Sex	Age	Diet Pattern	Mean± S.D	Group	Groups Compared	't'Value
M	15-20	Vegetarians	95.3 ± 4.77	(A)	A Vs B	3.17 **
		Non-vegetarians	114.1±16.7	(B)		
M	21-40	Vegetarians	97.54±4.5	(C)	C Vs D	3.65**
		Non-vegetarians	124.5± 18.8	(d)		
M	41-60	Vegetarians	94.6 ± 3.46	(E)	E Vs F	2.67 *
		Non-vegetarians	112.4 ± 16.25	(F)		
F	15-20	Vegetarians	95.5 ± 2.71	(A)	AVs B	2.671 *
		Non-vegetarians	106 ± 10.5	(B)		
F	21-40	Vegetatians	97.2 ± 1.98	(C)	C Vs D	4.75**
		Non vegetarians	129.6± 19.4	(D)		
F	41-60	Vegetarians	94.9 ± 2.962	(E)	E Vs F	2.02 N.S
		Non-vegetarians	108.0±17.08	(F)		

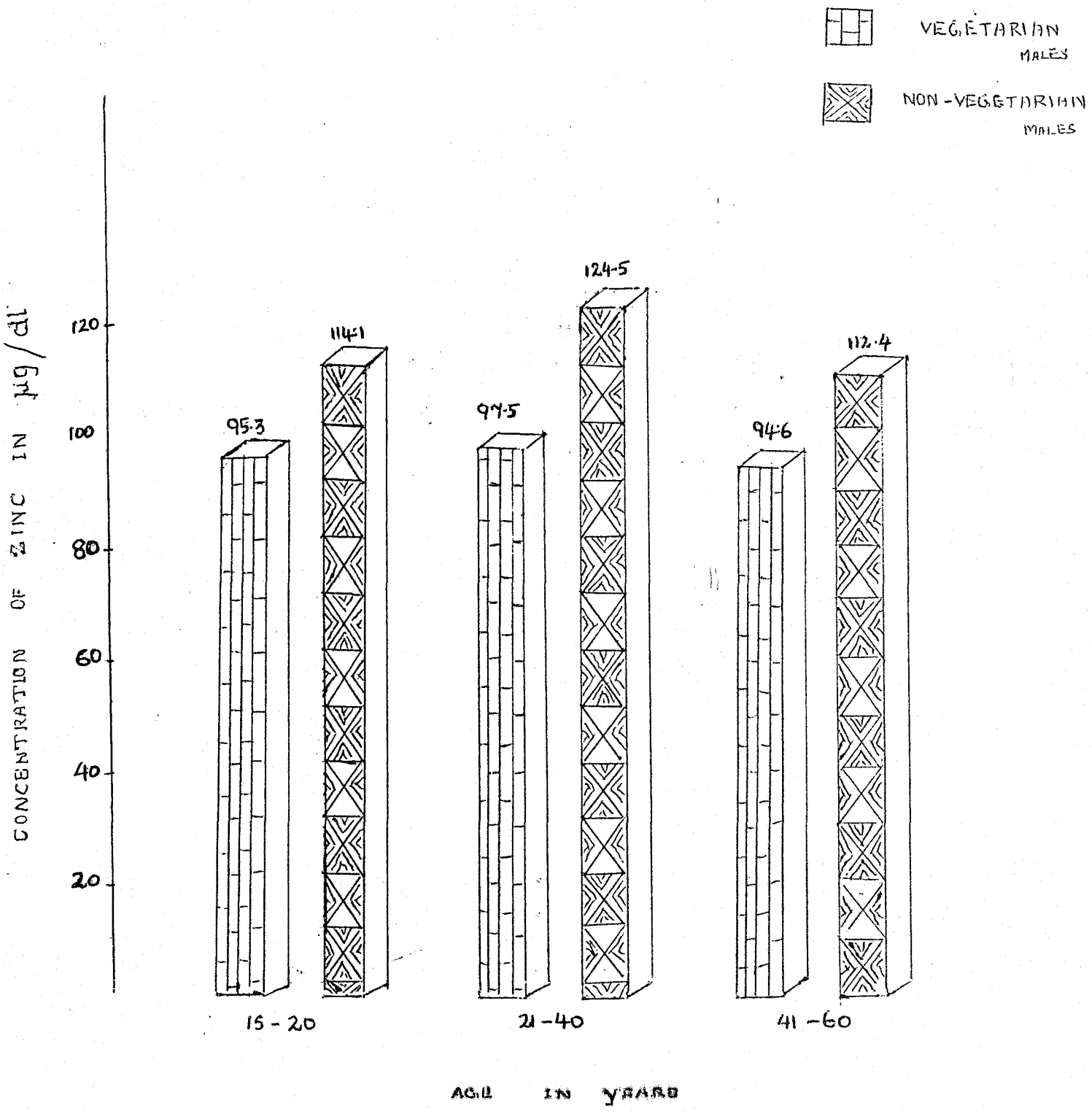
** Significant at 1 % level

* Significant at 5 % level

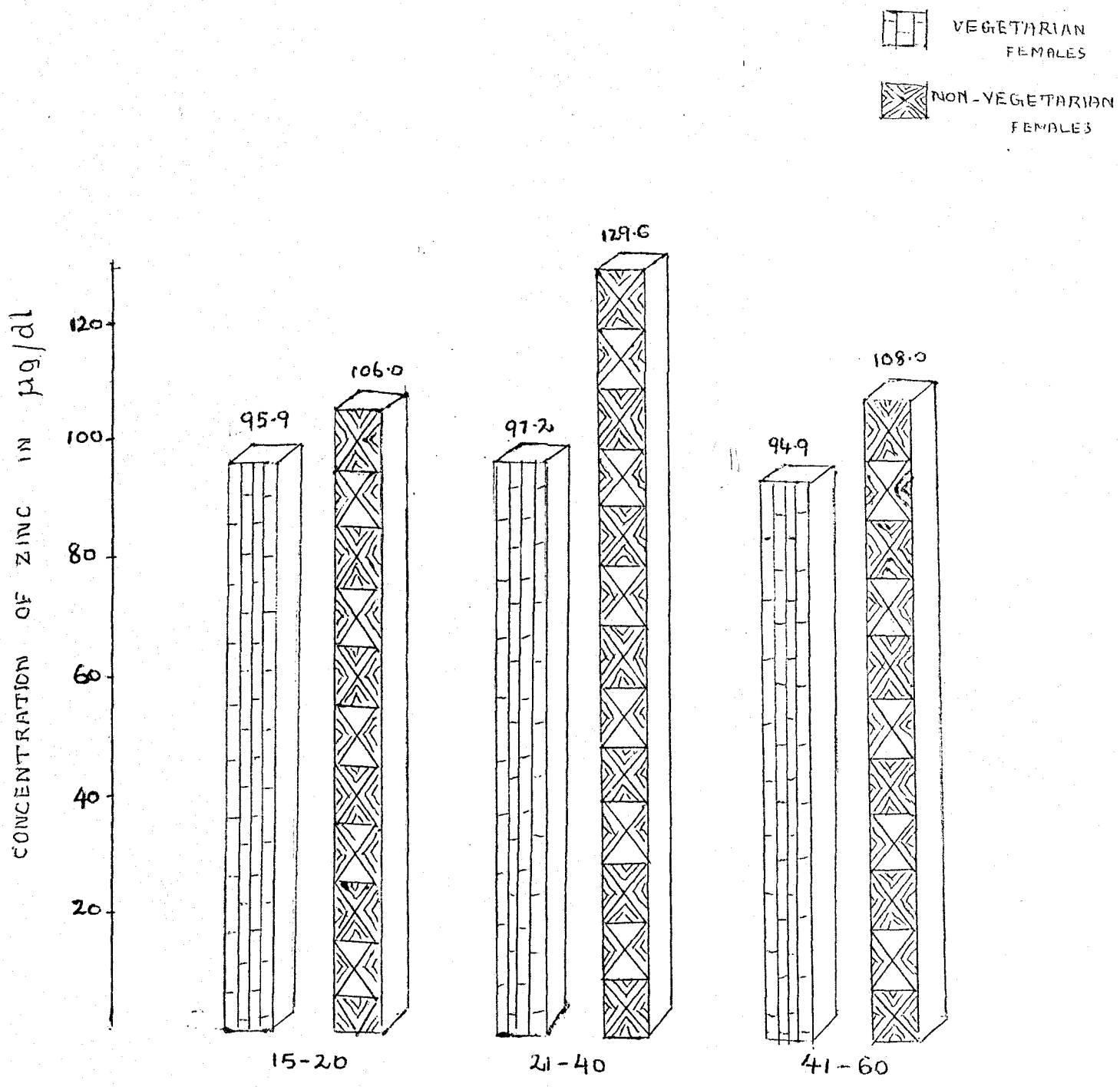
N.S. Not Significant.

It is seen from the values presented in Table-3, that in all age groups among men and women except in women of 41-60 years the non-vegetarians have heigher serum zinc content compared to the corresponding vegetarian group. The increase is statisfically

COMPARISON OF SERUM ZINC LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN MALES OF THE SAME AGE GROUP



COMPARISON OF SERUM ZINC LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN FEMALES OF THE SAME AGE GROUP



significant in men at one percent level in the age groups 15-20 and 21-40 and five percent level in the age group 41-60. Among women a significant rise at one percent level is observed in non-vegetarians of 21-40 age group and at five percent level in the age groups 15-20 while no change is noted in the age group 41-60.

The results indicate that non-vegetarian foods have a higher content of zinc in an absorbable form compared to vegetarian foods. A vegetarian gets all the essential minerals by taking cereals, millets and pulses in his diet. But the bioavailability of mineral such as zinc may be low in a total vegetarian diet because of the presence of substances such as phytic acid. Besides large amount of fibre may interfere with proper absorption. However, surveys conducted by NIN (1986) have shown that zinc deficiencies are no greater among vegetarians than non-vegetarians.

C. Effect of sex:

Table - 4 presents the comparision of serum zinc values between males and females of the same age group and same pattern of diet.

T A B L E - 4

Serum Zinc Levels in Males and Females in µg/dl

Diet Pattern	Age	Sex	Group	Mean ± S.D.	Groups Compared	't' Value
Vegetarians	15-20	M	(A)	95.3 ± 4.77	A Vs B	0.342N.S
		F	(B)	95.9 ± 2.71		
Vegetarians	21-40	M	(C)	97.54± 4.5	C Vs D	0.20N.S
		F	(D)	97.2 ± 1.98		
Vegetarians	41-60	M	(E)	94.6 ± 3.46	E Vs F	0.18N.S
		F	(F)	94.9± 2.962		
Non-vegetarians	15-20	M	(A)	114.1±16.7	A Vs B	0.111N.S
		F	(B)	106 ±10.5		
Non-vegetarians	21-40	M	(C)	124.5±18.8	C Vs D	0.642N.S
		F	(D)	129.6 +19.4		
Non-vegetarians	41-60	M	(E)	112.4±16.25	E Vs F	0.5 N.S
		F	(F)	108 + 17.08		

N.S. Not significant.

From table-4 it is clear that the mean serum zinc level is not changed significantly between the sexes of the same age group and same pattern of diet. Not only age but also sex does not influence the mean serum zinc values.

From the present study it is found that the serum zinc values in normal persons range between 80-150 µg/dl, the mean being 104.3±6.46 µg/dl. The earlier reported normal serum zinc value was 100(50-150µg/dl)(Tierney et al., 1986)

3. LEVELS OF SELENIUM IN SERUM:

(a) Effect of age:-

Table 5 presents the comparision of mean serum selenium among different age groups of the same sex and same pattern of diet.

T A B L E - 5

Serum Selenium levels in different age groups in µg/dl.

Diet Pattern	Sex	Age Group	Mean±SD	Groups compared.	't' Value
I. Vegetarians	M	15-20 (a_1)	10.94±0.97	a_1 Vs b_1	0.25 N.S.
	M	21-40 (b_1)	10.84± 0.81	a_1 Vs c_1	0.046 0.464 N.S
	M	41-60 (c_1)	10.92± 0.77	b_1 Vs c_1	0.213 N.S
	F	15-20 (x_1)	11.0±0.99	x_1 Vs y_1	0.867 N.S
	F	21-40 (y_1)	11.4±0.96	x_1 Vs z_1	0.214 N.S
	F	41-60 (z_1)	11.1±0.90	y_1 Vs z_1	0.622 N.S
II. Non-vegetarians	M	15-20 (a_2)	12.33±1.15	a_2 Vs b_2	1.659 N.S
	M	21-40 (b_2)	13.1 ±0.81	a_2 Vs c_2	0.467 N.S
	M	41-60 (c_2)	12.6±0.88	b_2 Vs c_2	1.12 N.S
	F	15-20 (x_2)	14.3±0.81	x_2 Vs y_2	2.0 *
	F	21-40 (y_2)	13.63±0.65	x_2 Vs z_2	0.155 N.S
	F	41-60 (z_2)	14.23±0.98	y_2 Vs z_2	1.652 N.S

N.S= Not significant * = Significant at 5% level.

From table 5 it is clear that the mean selenium level is not altered sifnificantly among different age groups of the same sex and same pattern of diet except in the case of non-vegetarian females of the age group 15-20 and 21-40 years.

(b) Effect of Diet:

Table-6 presents the comparison of serum selenium level between healthy vegetarians and non-vegetarians of corresponding age groups and sex.

T A B L E - 6

Serum Selenium levels in different patterns of diet in µg/dl.

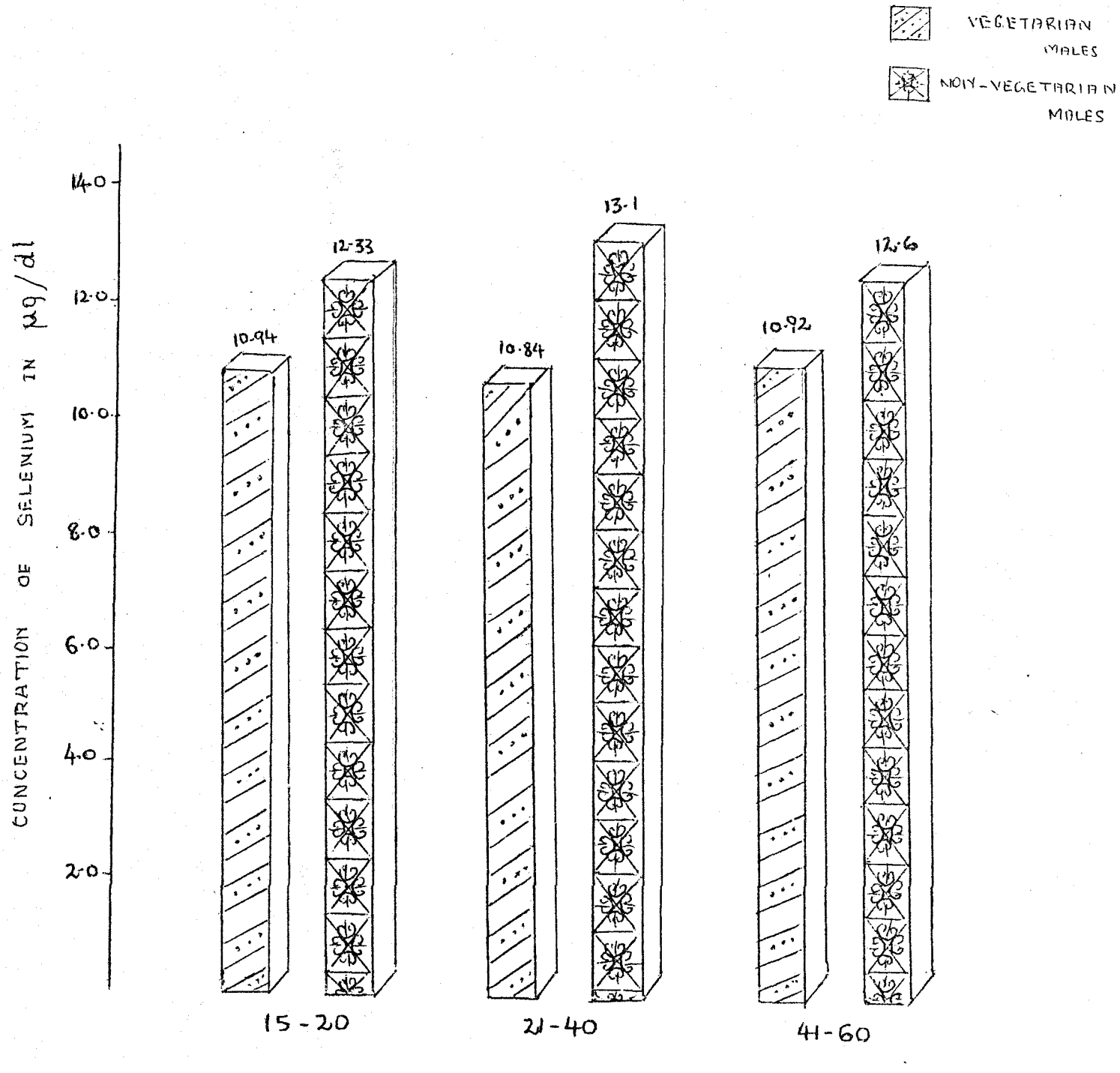
Sex	Age	Diet Pattern	Group	Mean ±S.D	Groups Compared	't' Value
M	15-20	Vegetarians	(A)	10.94±0.97	A Vs B	2.536*
		Non-vegetarians	(B)	12.33±1.15		
M	21-40	Vegetarians	(C)	10.84±0.81	C Vs D	6.569**
		Non-vegetarians	(D)	13.1± 0.81		
M	41-60	Vegetarians	(E)	10.92±0.77	E Vs F	3.818**
		Non-vegetarians	(F)	12.6± 0.88		
F	15-20	Vegetarians	(A)	11.0 ± 0.99	A Vs B	7.638**
		Non-vegetarians	(B)	14.3±0.81		
F	21-40	Vegetarians	(C)	11.4±0.96	C Vs D	6.04**
		Non-vegetarians	(D)	13.36±0.65		
F	41-60	Vegetarians	(E)	11.1 ±0.90	E Vs F	6.584**
		Non-vegetarians	(F)	14.23±0.98		

** Significant at 1 % level

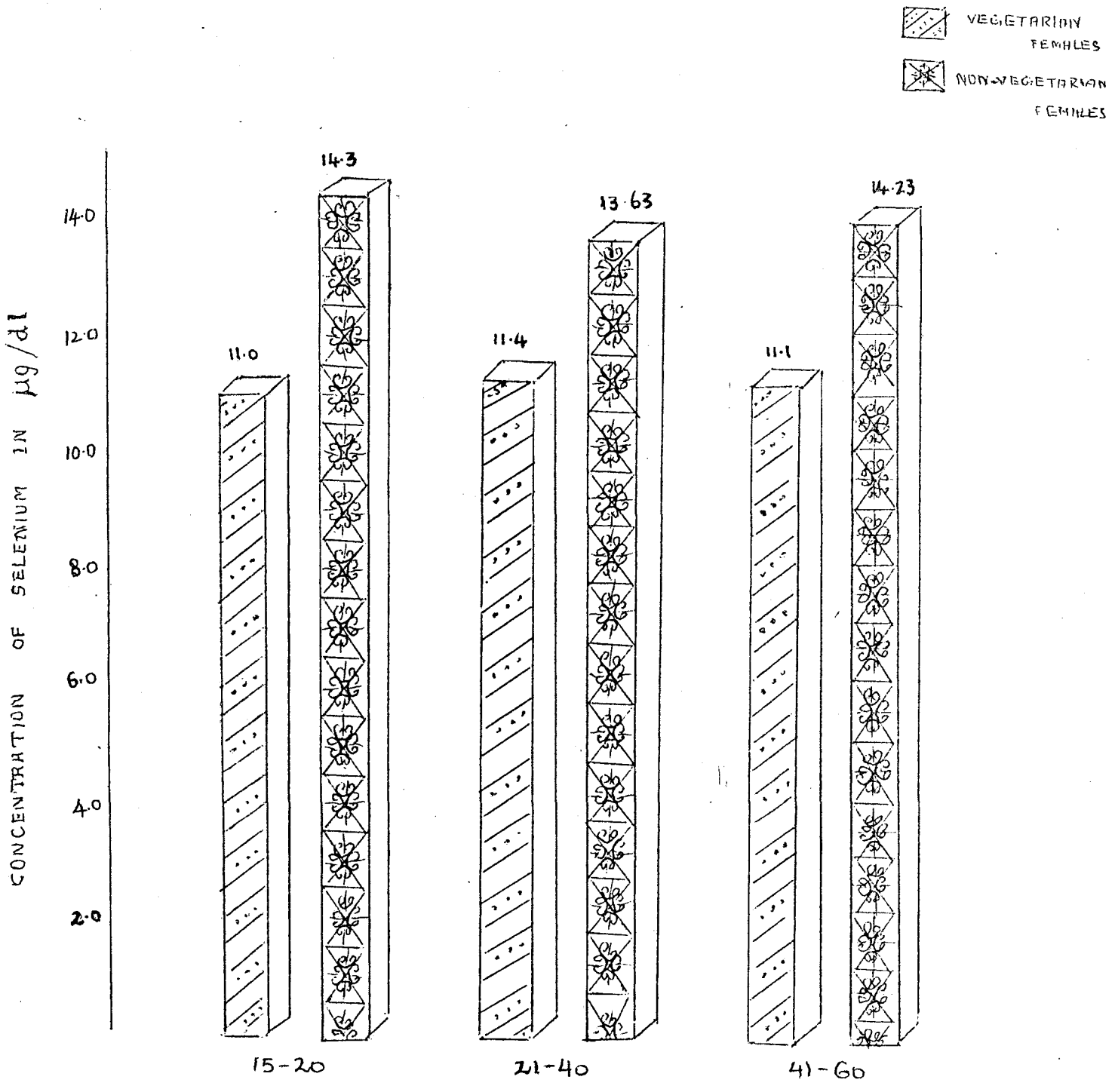
* Significant at 5% level.

It is seen from the values presented in Table-6 that in all age group among men and women, the non-vegetarians have higher serum selenium content compared to the corresponding vegetarian group. The increase is statistically significant

COMPARISON OF SERUM SELENIUM LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN MALES OF THE SAME AGE GROUP



COMPARISON OF SERUM SELENIUM LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN FEMALES OF THE SAME AGE GROUP



in men at one percent level in the age groups 21-40 and 41-60 years and five percent level in the age group 15-20. Among women significant rise at one percent level is observed in non-vegetarians of all age groups.

The results indicate that the type of food consumed could influence selenium levels in healthy persons.

Human serum selenium concentration is variable and depend in part on the selenium content and bioavailability in foods and water consumed (Terry et al., 1983)

The present finding that the serum selenium level of non vegetarians are significantly higher when compared to the vegetarians is due to the fact that animal foods are high in selenium while vegetables are poor sources of the same (Selvi, 1987).

C. Effect of Sex:

Table - 7 presents the comparison of serum selenium values between males and females of the same age group and same pattern of diet.

TABLE - 7

Serum Selenium levels in males and females in µg/dl

Diet Pattern	Age	Sex	Group	Mean ± S.D	Groups Compared	't' Value
Vegetarians	15-20	M	(A)	10.94±0.97	A Vs B	0.133NS
		F	(B)	11.0±0.99		
Vegetarians	21-40	M	(C)	10.84±0.81	C Vs D	1.346N.S
		F	(D)	11.4 ± 0.96		
Vegetarians	41-60	M	(E)	10.92± 0.77	E Vs F	0.417 N.S
		F	(F)	11.1 ± 0.90		
Non-vegetarians	15-20	M	(A)	12.33±1.15	A Vs B	3.766**
		F	(B)	14.3 ± 0.81		
Non-vegetarians	21-40	M	(C)	13.1 ± 0.81	C Vs D	1.76 N.S
		F	(D)	13.63± 0.65		
Non-vegetarians	41-60	M	(E)	12.6 ± 0.88	E Vs F	3.313**
		F	(F)	14.23± 0.98		

** Significant at 1% level

N.S.= Not significant.

From the table-7 it is clear that the difference in serum selenium level is not statistically significant between sexes of the same age group and same pattern of diet except in nonvegetarians of the age group 15-20 and 41-60 years. The serum selenium level of females are higher in the age groups 15-20 and 41-60 of non-vegetarian group. The difference is statistically significant at one percent level.

From the present investigation it is found that the serum selenium values in normal persons range between 9-14.8 µg/dl, the mean being 12.57 ± 0.905 µg/dl.

McConnel and Coworkers (1975) reported in healthy persons a normal mean selenium value of 11.8µg/dl (1975) which is slightly lower than the present value.

4. LEVELS OF CHROMIUM IN SERUM:

(a) Effect of age:

Table-8 presents the comparision of serum chromium among different age groups of the same sex and same pattern of diet.

T A B L E - 8

Serum chromium levels in different age groups in µg/dl.

DIET PATTERN	SEX	AGE	GROUP	MEAN ± S.D.	GROUPS COMPARED	't' VALUE
I. Vegetarians	M	15-20	(a_1)	0.69 ± 0.14	a_1 Vs b_1	0.025N.S
	M	21-40	(b_1)	0.68 ± 0.1	a_1 Vs c_1	0.85 N.S
	M	41.60	(c_1)	0.55 ± 0.33	b_1 Vs c_1	0.36 N.S
	F	15-20	(x_1)	0.62 ± 0.32	x_1 Vs y_1	0.87 N.S
	F	21-40	(y_1)	0.72 ± 0.08	x_1 Vs z_1	0.33 N.S
	F	41-60	(z_1)	0.66 ± 0.12	y_1 Vs z_1	1.153N.S
II.Non-Vegetarians:						
	M	15-20	(a_2)	0.83 ± 0.10	a_2 Vs b_2	1.25 N.S
	M	21-40	(b_2)	0.78 ± 0.07	a_2 Vs c_2	2.49 *
	M	41-60	(c_2)	0.7 ± 0.08	b_2 Vs c_2	2.22 *
	F	15-20	(x_2)	0.7 ± 0.06	x_2 Vs y_2	0.12 N.S
	F	21-40	(y_2)	0.71 ± 0.23	x_2 Vs z_2	1.25 N.S
	F	41-60	(z_2)	0.74 ± 0.07	y_2 Vs z_2	0.384 N.S

N.S. = Not Significant

* = Significant at 5% level.

From table - 8 it is clear that the difference in serum chromium level is not statistically significant among different age group of the same sex and same pattern of diet except among non-vegetarian males of the age groups 15-20 and 41-60 and 21-40 and 41-60 in which cases the differences are statistically signi-

(b) Effect of diet:

Table - 9 presents the comparision of serum chromium levels between vegetarians and nonvegetarians of the same sex and age groups.

T A B L E - 9

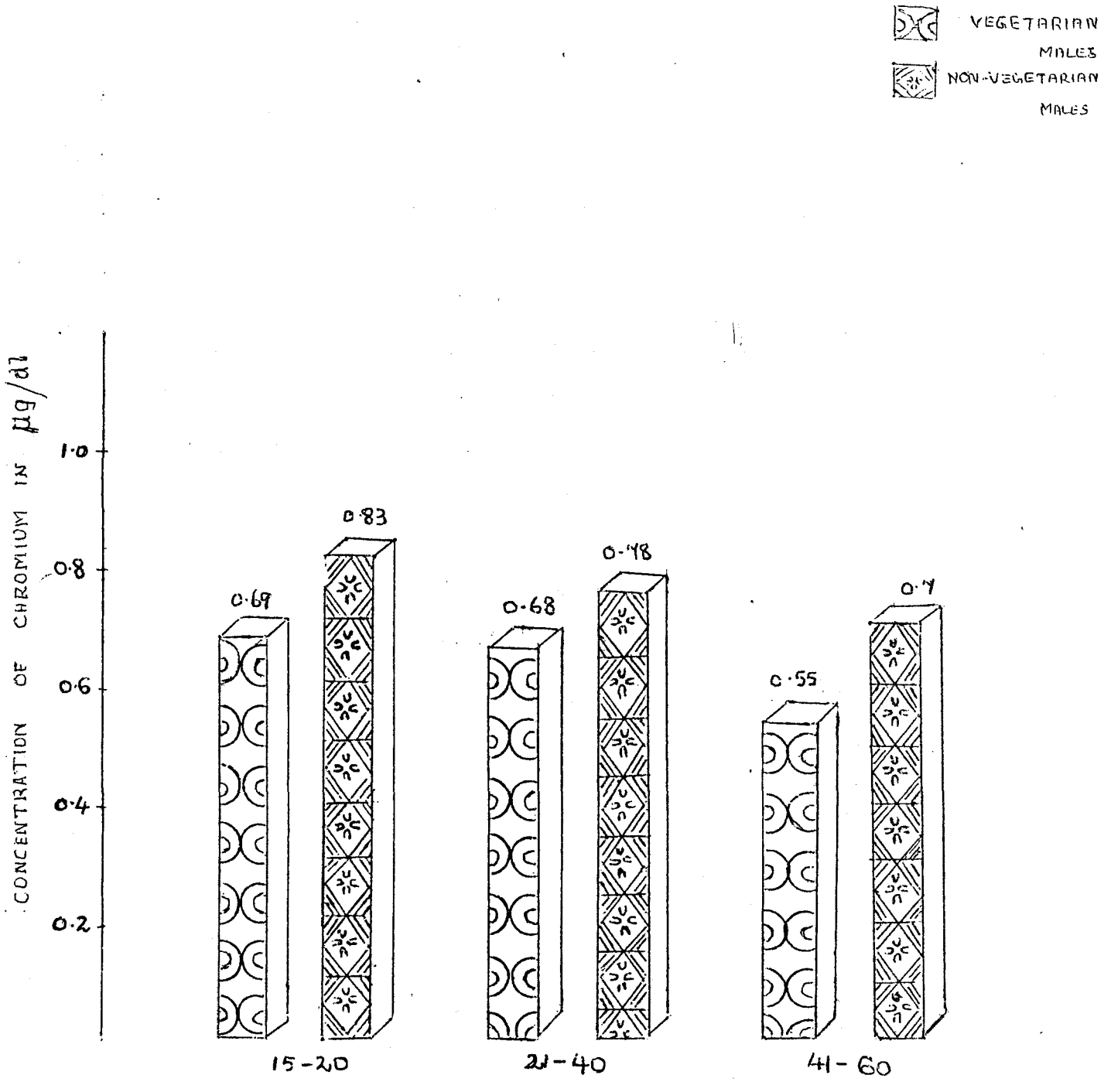
Serum chromium levels in different patterns of diet in µg/dl

Sex	Age	Diet Pattern	Group	Mean ± S.D.	Groups compared	't' Value
M	15-20	Vegetarians	(A)	0.69 ± 0.14	A Vs B	1.709N.S
		Non-vegetarians	(B)	0.83 ± 0.10		
M	21-40	Vegetarians	(C)	0.68 ± 0.1	C Vs D	0.347N.S
		Non-vegitarians	(D)	0.78 ± 0.07		
M	41-60	Vegetarians	(E)	0.55 ± 0.33	E Vs F	1.102N.S
		Non-vegetarians	(F)	0.7 ± 0.08		
F	15-20	Vegetarians	(A)	0.62 ± 0.32	A Vs B	0.70N.S
		Non-vegetarians	(B)	0.7 + 0.06		
F	21-40	Vegetarians	(C)	0.72 ± 0.08	C Vs D	0.12N.S
		Non-vegetarians	(D)	0.71 ± 0.23		
F	41-60	Vegetarians	(E)	0.66 ± 0.12	E Vs F	2.0N.S
		Non-vegetarians	(F)	0.74 ± 0.07		

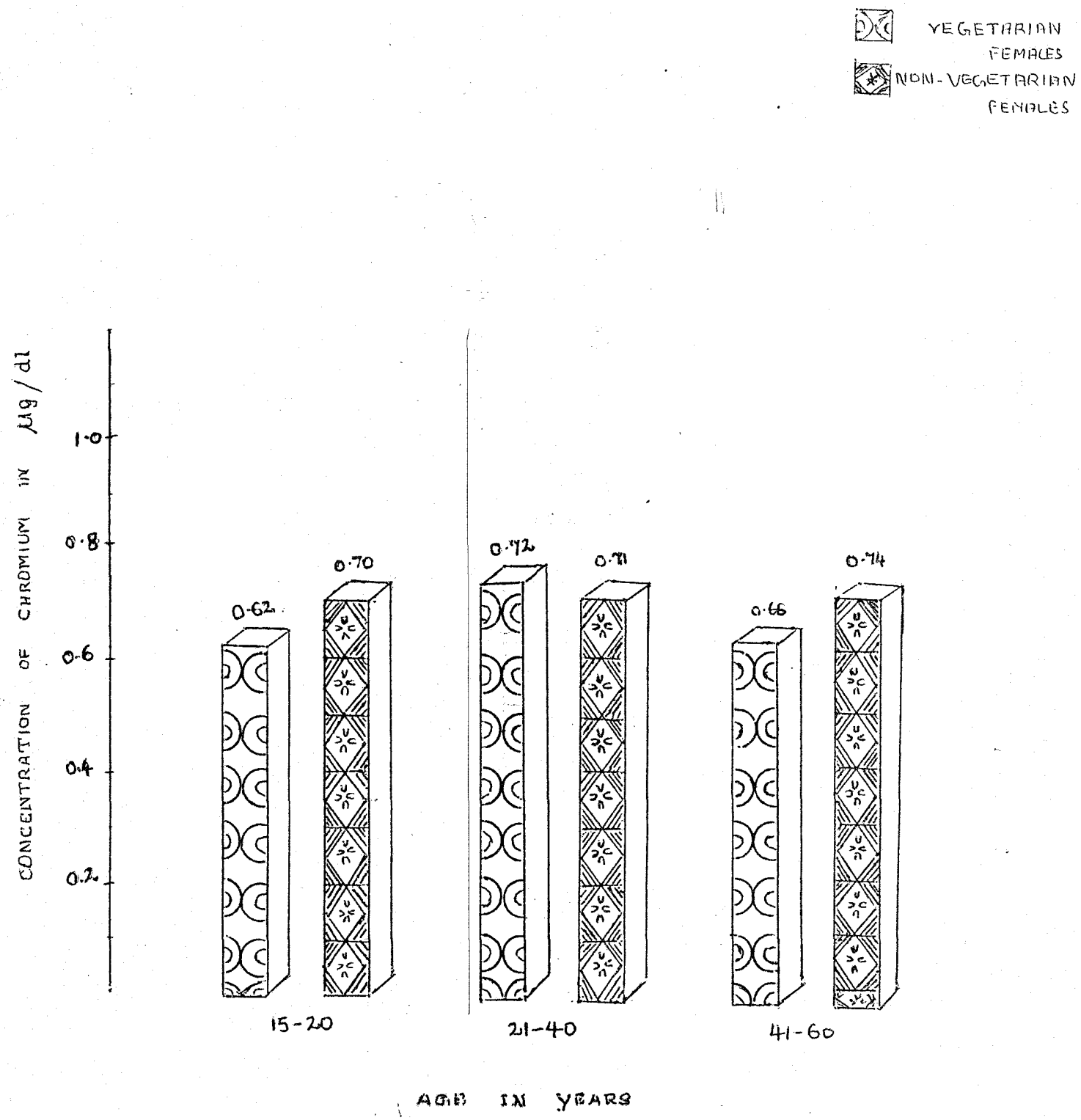
N.S. = Not significant.

It is seen from the values presented in Table-9 that the differences in serum chromium levels are not statistically sifnificant among vegetarians and non-vegetarians of corresponding sex and group.

COMPARISON OF SERUM CHROMIUM LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN MALES OF THE SAME AGE GROUP



COMPARISON OF SERUM CHROMIUM LEVELS BETWEEN VEGETARIAN AND NON-VEGETARIAN FEMALES OF THE SAME AGE GROUP



C. Effect of Sex:

Table - 10 presents the comparison serum chromium between males and females of the same age group and same pattern of diet.

TABLE - 10

Serum chromium levels in males and females in µg/dl.

Diet Pattern	Age	Sex	Group	Mean±S.D	Group Compared	't' Value
I Vegetarians	15-20	M	(A)	0.69±0.14	A Vs B	0.318N.S
		F	(B)	0.62±0.32		
	21-40	M	(C)	0.68±0.1	C Vs D	0.085N.S
		F	(D)	0.71±0.08		
	41-60	M	(E)	0.55±0.33	E Vs F	0.83N.S
		F	(F)	0.66±0.12		
II. Non-vegetarians	15-20	M	(A)	0.83±0.10	A Vs B	3.020**
		F	(B)	0.7±0.06		
	21-40	M	(C)	0.78±0.07	C Vs D	1.014N.S
		F	(D)	0.71±0.23		
	41-60	M	(E)	0.7 ±0.08	E Vs F	1.052N.S
		F	(F)	0.74±0.07		

** Significant at 1% level

N.S. = Not significant

From the above table it is clear that serum chromium is not statistically significant between sexes of the same age group and same pattern of diet except in nonvegetarian group consisting of males and females of the age group 15-20 which is significant at one percent level.

From the values obtained in the present study, it is found that the serum chromium level in normal persons range between 0.5-1.0µg/dl the mean being 0.695±0.05 µg/dl.

The earlier reported normal serum chromium values were 1.67µg/dl (Liu and Morris,1978), 1.62µg/dl pekkarek et al., 1974). 0.73µg/dl (Grafflege et al., 1974; Seeling et al., 1975).

Summary and Conclusion

V. SUMMARY AND CONCLUSION

The results obtained in the study Serum Zinc, Selenium and chromium levels in normal persons are summarised as follows;-

A group of one hundred and sixteen normal healthy persons of age groups 15-20, 21-40 and 41-60 years, both sexes and vegetarian and nonvegetarian patterns of diet, were selected and blood samples were obtained from them to estimate serum zinc, selenium, and chromium levels.

Levels of zinc in serum:

In vegetarian males of different age groups the serum zinc values were 15-20: 95.3 ± 4.77 µg/dl; 21-40: 97.54 ± 4.5 µg/dl; and 41-60: 94.6 ± 3.46 µg/dl.

There was no statistically significant change in serum zinc values among different age groups of vegetarian men.

In non vegetarian males of different age groups the zinc values were 15-20: 114.1 ± 16.7 µg/dl; 21-40: 124.5 ± 18.8 µg/dl; 41-60: 112.4 ± 16.25 µg/dl.

There was no statistically significant change in serum zinc values among non-vegetarian males of different age groups.

Hence, it is concluded that age has no effect on serum zinc level of men consuming similar diet.

The above results also show that non-vegetarian males had higher serum zinc content compared to the corresponding vegetarian group. The increase was statistically significant in men at one percent level in the age groups 15-20 and 21-40 and at five percent level in the age group 41-60 years.

In vegetarian females of different age groups the serum zinc values were as follows: 15-20: 95.9 ± 2.7μg/dl 21-40: 97.2 ± 1.98 μg/dl; 41-60: 94.9 ± 2.962 μg/dl. There was no statistically significant change in serum zinc values among different age groups of vegetarian women.

In non vegetarian females of different age groups the serum zinc values were 15-20:106.0± 10.5 μg/dl; 21-40 129.6 ± 19.4 μg/dl and 41-60: 108.0 ± 17.08 μg/dl. The highest serum zinc level was in the age group 21-40 which differed significantly from the values of the other two groups. It is not clear why this increase is found in non-vegetarian women of 21-40 years.

It is further observed that nonvegetarian females had higher serum zinc content compared to the corresponding vegetarian age group, the increase being statistically significant at one percent level in the age group 21-40 years and five percent level in the age group 15-20 years. However there was no statistically significant change in the age group 41-60 years.

Serum zinc levels when compared between males and females of the same age group and same pattern of diet, showed no significant change between the sexes.

From the above results it is clear that not only age but also sex did not influence very much the mean serum zinc values whereas, diet plays an important role in maintaining serum zinc levels in normal persons.

From the present study it is found that the serum zinc values in normal persons range between 80-150 µg/dl the mean being 104.33 ± 6.46 µg/dl. The earlier reported normal serum zinc value was 100(50-150 µg/dl) (Tierney et al., 1986).

Levels of selenium in serum:

In vegetarian males of different age groups the serum Selenium values were found to be as follows: 15-20: 10.94 ± 0.97 µg/dl; 21-40: 10.84 ± 0.81 µg/dl; and 41-60: 10.92 ± 0.77 µg/dl. The mean selenium levels were not altered significantly among different age groups of vegetarian men.

In non-vegetarian males of different age groups serum selenium values were: 15-20: 12.33 ± 1.15 µg/dl; 21-40: 13.1 ± 0.81 µg/dl; 41-60: 12.6 ± 0.88 µg/dl. There was no statistically significant change in serum selenium values among non-vegetarian males of different age groups.

Hence, it is concluded that age has no effect on serum selenium level of men.

From the above results it is also clear that non-vegetarian males of different age groups have a higher serum selenium level compared to the corresponding vegetarian group. This increase is statistically significant at one percent level in the age groups 21-40 and 41-60 years and at five percent level in the age group 15-20 years.

In vegetarian females of different age groups the serum selenium values were: 15-20: 11.0 ± 0.99 µg/dl; 21-40: 11.4 ± 0.96 µg/dl; 41-60: 11.1 ± 0.90 µg/dl. There was no statistically significant change in serum selenium values among age groups of vegetarian women.

In non-vegetarian females of different age groups the serum selenium values were: 15-20: 14.3 ± 0.81 µg/dl; 21-40: 13.63 ± 0.65 µg/dl; 41-60: 14.28 ± 0.98 µg/dl. There was no significant change in the serum selenium values of non vegetarian women when age groups 15-20 and 41-60, 21-40 and 41-60 were compared.

From the above results it is clear that the non-vegetarian females had increased serum selenium levels when compared with the corresponding vegetarian female ~~when compared with the corresponding vegetarian female~~ group. The increase was statistically significant at

one percent level in all age groups.

The difference in Serum selenium levels was not statistically significant between sexes of the same age group and same pattern of diet except in non-vegetarians of the age group 15-20 and 41-60 years in which cases the differences were significant at one percent level.

The results indicate that the type of food consumed influences selenium levels in healthy persons. Human serum selenium concentration is variable and depand in part on the selenium content and bioavailability in foods and water consumed (Terry et al., 1983).

From the present investigation it is found the serum selenium values in normal persons range between 9-14.8 µg/dl, the mean being 12.57 ± 0.905 µg/dl. Mcconnel and co workers (1975) reported in healthy persons a normal mean serum selenium value of 11.8 µg/dl (1975) which is slightly lower than the present value.

Levels of chromium in serum:

In vegetarian males of different age groups the serum chromium values were as follows: 15-20: 0.69 ± 0.14 µg/dl; 21-40: 0.68 ± 0.1 µg/dl; 41-60: 0.55±0.33µg/dl. There was no statistically significant change in serum chromium values among different age groups of vegetarian men.

In non-vegetarian males of different age groups the serum chromium values were: 15-20: 0.83 ± 0.10 µg/dl; 21-40: 0.78 ± 0.07 µg/dl; 41-60: 0.7 ± 0.08 µg/dl. There was significant difference at five percent level in the serum chromium levels between the age groups 15-20 and 41-60, 21-40 and 41-60. The mean chromium value decreased with increase in age.

In vegetarian females of different age groups the serum chromium values were: 15-20: 0.62 ± 0.32 µg/dl; 21-40: 0.78 ± 0.08 µg/dl; 41-60: 0.66 ± 0.12 µg/dl. There was no statistically significant change in serum chromium values among different age groups of vegetarian women.

In non-vegetarian females of different age groups the serum chromium levels were 15-20: 0.7 ± 0.06 µg/dl; 21-40: 0.7 ± 0.23 µg/dl; 41-60: 0.74 ± 0.07 µg/dl. There was no statistically significant change in serum chromium values among different age groups of nonvegetarian women.

It is seen from the chromium values presented, that there was no significant difference when vegetarian males and females were compared with the corresponding non vegetarian group. Hence it is clear that the diet pattern has no effect on the level of serum chromium in both men and women. This is due to the fact that there is no

great difference between the chromium content of vegetable and animal foods (Vide P= 117).

From the values obtained in the present study it is found that the serum chromium level in normal persons range between 0.5-1.0 µg/dl the mean being 0.695 ± 0.05µg/dl

The earlier reported normal serum chromium values were 1.67 µg/dl(Liu and Morris, 1978). 1.62µg/dl (Pekarek et al., 1974) 0.73 µg/dl (Grafflage et al., 1974) (Sealing et al., 1975).

To conclude, serum zinc and selenium levels are greatly influenced by diet. Non vegetarian diet enhance the serum levels of both zinc and selenium. Chromium levels are not affected by diet vegetarian or non-vegetarian.

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