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# Relationships among Age, Physical Measurements and Protein Intake and Metabolism in Older Adult Female Vegetarians and Nonvegetarians

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To the Graduate Council:

I am submitting herewith a dissertation written by Mary Elizabeth Kunkel entitled "Relationships among Age, Physical Measurements and Protein Intake and Metabolism in Older Adult Female Vegetarians and Nonvegetarians." I have examined the final electronic copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Human Ecology.

Roy E. Beauchene, Major Professor

We have read this dissertation and recommend its acceptance:

Rossie L. Mason, Ada Marie Campbell, James N. Liles

Accepted for the Council:
Dixie L. Thompson

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

# RELATIONSHIPS AMONG AGE, PHYSICAL MEASUREMENTS AND PROTEIN INTAKE AND METABOLISM IN OLDER ADULT FEMALE VEGETARIANS AND NONVEGETARIANS

A Dissertation

Presented for the

Doctor of Philosophy

Degree

The University of Tennessee, Knoxville

Mary Elizabeth Kunkel

August 1979

To the Graduate Council:

I am submitting herewith a dissertation written by Mary Elizabeth Kunkel entitled "Relationships among Age, Physical Measurements and Protein Intake and Metabolism in Older Adult Female Vegetarians and Nonvegetarians." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Home Economics.

Roy E. Beauchene

Roy E. Beauchene, Major Professor

We have read this dissertation
and recommend its acceptance:

Ronnie L. Mason

Ada Marie Campbell

James M. Gilbert

Accepted for the Council:

L. Erwin Rosh

Vice Chancellor
Graduate Studies and Research

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ABSTRACT

This study concerned the relationships among age, physical measurements, protein intake and the urinary excretion of protein-derived matabolites in 125 adult female vegetarians and nonvegetarians. The vegetarian group (AV) included 57 lactoovovegetarians (LOV) and 6 vegans (V). The remaining 62 subjects were nonvegetarians (NV). The subjects ranged in age from 40 to 92 years. The AV were primarily Seventh-Day Adventists who had been recruited through their church groups. The NV were recruited primarily through Jesus Christ of Latter Day Saints and United Methodist church groups as well as Extension Homemakers Clubs. Measurements of height, weight and triceps skinfold thickness, a 7-day dietary record and a 24-hour urine sample were obtained from each subject. Mean daily intakes of energy, total protein, animal protein and vegetable protein were calculated from the dietary records. The urine samples were analyzed for total nitrogen, urea nitrogen, ammonia nitrogen, creatinine, hydroxyproline and inorganic sulfate. Data were adjusted to the mean age of the sample (59.4 years).

Mean (± SEM) heights of the V, LOV and NV groups did not differ and were 162.8 ± 2.4, 162.0 ± 0.8 and 161.1 ± 0.8 cm, respectively. The V (51.0 ± 5.6 kg) weighed significantly less and had a significantly smaller lean body mass (LBM) (39.9 ± 2.1 kg) than LOV (65.8 ± 1.8 and 43.9 ± 0.7 kg, respectively) and NV (66.7 ± 1.6 and 43.7 ± 0.6 kg, respectively). Triceps skinfold thickness and percent body fat for V (19.8 ± 3.6 mm and 14.8 ± 3.9%) were less than those of LOV (28.5 ± 1.1 mm

and 24.1 ± 1.2%). Both vegetarian groups had smaller skinfold thicknesses and percent body fats than NV (32.4 ± 1.0 mm and 28.1 ± 1.1%).

Mean energy intakes for the groups did not differ significantly and were all less than the NRC-RDA. The V consumed 1601 ± 133; the LOV, 1509 ± 42; and the NV, 1524 ± 42 kcal/day. The LOV (54.6 ± 1.3 g) and V (52.2 ± 4.2 g) consumed comparable mean daily amounts of total protein which were significantly less than that consumed by the NV (66.5 ± 1.6 g). The NV consumed 45.8 ± 1.7 g or 68.2 ± 2.2% of total protein from animal sources which was significantly more than the 16.0 ± 1.7 g and 29.5 ± 2.1% of total protein from animal sources consumed by LOV. The V consumed 38.2 ± 3.2 g or 74.8 ± 5.0% of total protein from vegetable sources which was significantly more than the 28.2 ± 1.1 g or 52.1 ± 1.7% of total protein as vegetable protein consumed by the LOV. Both vegetarian groups consumed significantly more vegetable protein than the NV (10.8 ± 1.1 g or 16.4 ± 1.8% of total protein).

There were no significant differences in urinary total nitrogen excretion for the V, LOV and NV groups. The values were 8.50 ± 1.10, 8.49 ± 0.35 and 9.00 ± 0.36 g/day, respectively. Urea nitrogen excretion did not differ significantly; V excreted 7.08 ± 1.09 g; the LOV, 7.24 ± 0.36 g; and the NV, 7.77 ± 0.36 g/day. The LOV excreted 174.6 ± 13.4 mg of ammonia nitrogen/day which was significantly less than the 226.6 ± 15.1 mg excreted by the NV. The 201.2 ± 42.0 mg of ammonia nitrogen excreted by the V did not differ significantly from that of either other group. The creatinine excretions of groups V and LOV were 1.30 ± 0.14 and 1.33 ± 0.04 g/day, respectively, and were significantly less than the

1.54 ± 0.05 g excreted by the NV. Hydroxyproline (HOP) excretion by the V (28.3 ± 4.8 mg/day) was significantly less than the 35.5 ± 1.7 mg excreted by the NV. The 33.2 ± 1.6 mg of HOP/day excreted by the LOV did not differ significantly from that of either other group. Inorganic sulfate excretion did not differ among the groups V, LOV and NV; the values were 1.41 ± 0.22, 1.55 ± 0.07 and 1.57 ± 0.07 g/day, respectively.

Height decreased linearly with age in both the AV and NV groups. Weight and LBM decreased with age in NV; in the AV, the variables increased through age 65 and decreased thereafter. Skinfold thickness and percent body fat were significantly different between the AV and NV and tended to decrease with age in both groups. Energy and protein intakes in the AV decreased almost linearly with age; intakes of the NV increased until the sixth decade and decreased thereafter. Total nitrogen excretion showed approximately the same relationship to age as protein and energy intakes. Ammonia nitrogen and creatinine excretions were significantly different between the AV and NV. Urinary excretions of urea, HOP and inorganic sulfate did not differ between the AV and NV nor did they show a relationship to age.

The most notable differences observed between the AV and NV subjects were in body fat (skinfold thickness), protein intake and urinary creatinine. The 2 groups were similar in height and urinary HOP and inorganic sulfate throughout the age span studied. Regressions of body weight, LBM and body fat on age tended to follow a similar pattern within each group. Energy intake, protein intake and urinary total nitrogen also showed similar relationships to age within each group. The V

subjects differed from the NV subjects with respect to body weight, body fat, LBM and urinary HOP and differed from the LOV with respect to body weight, body fat and LBM. It can be concluded that the consumption of a vegan diet may have an effect on selected physical measurements and the intake and metabolism of protein as compared to a nonvegetarian diet while the effect of consumption of a lactoovovegetarian diet is minimal.

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# CHAPTER I

## INTRODUCTION

Knowledge of the relationships between the dietary protein requirement and aging is, at best, limited. Protein metabolic status of any age group is affected by the nature of the long-term dietary intake, body protein mass and other conditions which affect the body's utilization of protein such as various disease states and changes in absorptive and excretory ability. Assessing protein status in older people is further complicated by the fact that these individuals represent the accumulation of a wide variety of physical, environmental and dietary influences.

One of the major factors affecting protein metabolic status is the quality and quantity of protein intake. Therefore, in studying the effects of long-term consumption of protein with different amino acid composition, it would be advantageous to utilize subjects normally consuming such proteins, e.g., vegetarians and nonvegetarians. The variations in quality and quantity of the proteins consumed by vegetarians and nonvegetarians may result in differences in protein utilization and subsequently in protein metabolic status.

Another influence on protein metabolic status is the total body protein mass. The aging process is generally accompanied by decreases in muscle mass, changes in collagen structure and changes in other components of the body protein mass.

One method of assessing protein metabolic status involves measuring urinary excretion of metabolites derived either directly or indirectly from dietary protein. Among the metabolites that are considered to be directly related to the quality and/or quantity of the dietary protein are total nitrogen, urea, ammonia and inorganic sulfate. Among the urinary metabolites that are indirectly related to dietary protein intake are creatinine and hydroxyproline. In individuals consuming diets containing minimal amounts of creatinine and hydroxyproline, such as vegetarians, urinary excretion of these compounds may be considered to reflect endogenous protein metabolism.

It was the purpose of this study to examine the relationships among age, physical measurements, the dietary intake of protein and the excretion of protein-derived metabolites in older adult female vegetarians and nonvegetarians.

# CHAPTER II

## REVIEW OF LITERATURE

In view of the growing numbers of elderly in the population, it is increasingly important to investigate the adequacy of the diets consumed by this segment of society. The aging process is characterized by progressive changes in various physiological systems leading to decreased functional ability (1). These physiological decrements have implications in terms of nutrient requirements and hence, nutritional status. However, actual dietary requirements for the elderly as a separate segment of the adult population have not been defined. Instead, it has been assumed that by and large there are little or no significant changes in nutrient needs accompanying aging (2).

### I. PROTEIN REQUIREMENTS AND INTAKES

Recently Scrimshaw et al. (3) assessed protein requirements in older women and compared their values to those obtained by Bricker and Smith (4) using young women. Obligatory urinary nitrogen losses per unit body cell mass were significantly greater for older than for younger women, even though total protein requirements did not differ between the 2 groups. In a similar study comparing young and old men, Uauy et al. (5) confirmed the results of Scrimshaw et al. (3). Nitrogen losses per unit of body weight were higher for both old and young males than for old females. Obligatory urinary nitrogen excretion per unit of body cell mass was significantly higher for old males than for young males, but

did not differ from that for old female subjects. In addition, body cell mass per unit of creatinine excretion was higher for old subjects, reflecting the decrease in muscle mass with aging. Zanni et al. (6) found endogenous urinary nitrogen excretion per unit of body weight to be less for old than for young males (7) but the same when expressed in relation to lean body mass, to basal metabolic rate or to urinary creatinine. Uauy et al. (5) interpreted their findings as indicating either a selective loss of lean body mass with the visceral tissue being preserved, thus contributing a greater proportion to total body protein synthesis or a decreased ability to adapt to a protein-free diet with increasing age. Zanni et al. (6), in contrast, reported their old men retained lean body mass of uniform composition and that the elderly have a higher total protein requirement than the younger men due to a decreased efficiency of protein utilization. Cheng et al. (8) reported that protein digestibility and the ability to adapt to changes in protein intake between young and older adult males did not significantly differ.

While the total amount of protein required by the elderly may not differ from that required by younger adults, optimum amino acid balance of the diet has not been established. Using the nitrogen balance technique, Tuttle, Swendseid and colleagues (9) found that elderly subjects have a higher requirement for 1 or more of the indispensable amino acids, e.g., methionine, and that the requirement for these amino acids may be related both to the total dietary nitrogen intake and to the source of the "nonspecific nitrogen." In contrast to these findings,

Watts et al. (10) reported achievement of nitrogen balance in older men with methionine levels similar to those required for younger subjects. The reasons for these apparent contradictions have not been elucidated. Using a method based on plasma amino acid concentrations, Young and coworkers (1, 11) have conducted a limited number of studies to assess amino acid needs of the elderly. While certainly not conclusive, their results seem to indicate that the indispensable amino acid requirements of the elderly may be greater than those of younger adults per unit of body cell mass.

The adult dietary requirement for protein consists of an amount needed to replace endogenous nitrogen losses corrected for the biological quality of the protein eaten (1). Using 0.47 g protein per kg of body weight as the minimum endogenous nitrogen output and allowing for a 30% variation and a 70% net protein utilization, the National Research Council (12) has set its Recommended Dietary Allowances (RDA) at 0.6 g high quality or 0.8 g mixed protein per kg of body weight per day throughout adulthood. Scrimshaw et al. (3) and Uauy et al. (5) calculated "safe" levels of intake for egg and milk protein in older men and women as 0.55 and 0.42 g of protein per kg of body weight per day, respectively. Zanni et al. (6) calculated a protein requirement for older men of 0.51 g of egg protein per kg of body weight per day. Both estimates are less than those of either the FAO/WHO (13) or NRC (12).

Results of the USDA Household Food Consumption Survey (14) using 0.9 g of protein per kg of body weight as the standard and the Health and Nutrition Examination Survey (HANES) (15) using 1 g of protein per kg of

body weight as the standard indicated inadequate intakes of protein in the elderly. Jansen and Harrill (16), Greger and Sciscoe (17), Jordan (18), Kohrs et al. (19) and MacLennan et al. (20) reported mean protein intakes that met or exceeded 0.8 g per kg of body weight. Justice et al. (21) found one-third of their subjects and Jordan (18) reported two-fifths of her subjects consumed less than the RDA (12). The Nutrition Canada Survey (22) results indicated that approximately 40% of all females and the same percentage of Indian males had protein intakes of less than 0.7 g per kg of body weight. The Ten-State Nutrition Survey (23) data indicated mean intakes of protein for low income black males and several groups of females were below 1 g per kg of body weight, but all groups consumed greater than 0.8 g per kg of body weight. DaCosta and Moorhouse (24) reported a range of protein intakes (adjusted to a reference height of 155 cm) in elderly women of 33 to 60 g per day and concluded that low protein intakes in the elderly did not have an adverse effect on body protein content.

## II. ENERGY REQUIREMENTS AND INTAKES

Energy requirements should reflect individual differences in energy expenditure so that a state of balance can be maintained. Harper (25) pointed out that energy intake of the elderly must be adjusted downward to compensate for the decreases in lean body mass, resting metabolic rate and physical activity accompanying aging. The NRC-RDA (12) for persons greater than 50 years of age is 2400 kcal per day for men and 1800 kcal per day for women. These values assume light to sedentary activity and must be adapted to the individual.

Results of the major studies including HANES (15), the Ten-State Nutrition Survey (23), the USDA Household Food Consumption Survey (14) and the Missouri Food Consumption Survey (19) indicated mean energy intakes less than the standards used for the elderly. Inadequate mean energy intakes were also reported by Davidson et al. (26), Justice et al. (21), McGandy et al. (27), Harrill et al. (28), Thompson,[1] and Greger and Sciscoe (17). Macleod et al. (29) reported adequate energy intakes that remained constant with age when expressed in relation to body weight of the elderly.

## III. VEGETARIAN DIETS

Vegetarian diets may be based on plant sources only (vegan), plant plus dairy products (lactovegetarian) or plant plus dairy and egg products (lactoovovegetarian). As the restrictiveness of the diet decreases, the likelihood of nutritional inadequacy also decreases, so that the lactoovovegetarian diet has been generally regarded as adequate in nutrient intake. Hardinge and Stare (30) found that intakes of nutrients by lactoovovegetarians and vegans met or exceeded the RDA with the exception of vitamin $B_{12}$ which was low in the vegan diet. In the same study, it was noted that almost half of both the male and female adult lactoovovegetarians and nonvegetarians consumed less than the RDA for energy. Brown and Bergan (31) reported mean caloric intakes for 23

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[1] G. S. Thompson, "The Effects of Dietary Supplements on Bone Density and Nutritional Status of Elderly Women" (unpublished Ph.D. thesis, The University of Tennessee, Knoxville, 1973).

to 50 year old male and female vegetarians that were 59 and 70%, respectively, of the RDAs for those age and sex groups.

A major concern of the vegetarian diet is the protein quality and quantity. Plant foods are known to be limiting in 1 or more of the indispensable amino acids thus necessitating use of "complementary" proteins to provide proteins of a quality comparable to those of animal foods. The protein adequacy of a vegetarian diet can also be affected by the low energy intake of many vegetarians which would necessitate the use of protein as an energy source. Register and Sonnenberg (32) discussed data on protein quality and quantity of the vegetarian diet and concluded that both were adequate.

Hardinge et al. (33) analyzed amino acid intakes of lactoovovegetarians and vegans and found that intakes of all groups ranged from more than twice to many times the minimum indispensable amino acid requirement. Lin et al. (34) reported that vegans could utilize ingested protein slightly more efficiently than subjects on mixed protein diets. Nitrogen balance studies showed that although the vegans consumed less nitrogen, they were either more positively or less negatively in balance than the mixed diet subjects on low protein diets. These results were confirmed by Register et al. (35) who concluded that at a level of 10 g of nitrogen per day, there was no significant difference in the ability of young adult humans to maintain nitrogen balance when on a vegan, lactoovovegetarian or nonvegetarian diet.

## IV. LEAN BODY MASS

Lean body mass (LBM) has been found to peak in the third decade of life, to fall slowly for the next 2 decades, and then to decrease more rapidly (36). Forbes and Reina (37) found that by age 65 to 70 years the average male has 12 kg less LBM than at age 25; the average female has 5 kg less.

If LBM represents metabolically active tissue, decreases in LBM would be expected to have obvious implications in terms of protein and energy requirements (1). The major physical measurement related to LBM appears to be body height. Forbes (38) and Hume (39) both found that LBM could be best predicted by a regression equation incorporating the cube of height. Lean body mass has been found to be very highly correlated with urinary creatinine excretion (40). This finding reflects the fact that creatine, the major physiological precursor of creatinine, is found principally in muscle. Therefore, body creatine, which constitutes a constant percentage of the LBM, and muscle protein turnover are related to urinary creatinine excretion (40).

## V. BODY FAT

It is generally recognized that body fat increases with age in conjunction with the decrease in LBM (41). An indirect method of estimating body fat involves measurement of skinfold thickness. Application of this technique requires the assumption that subcutaneous fat constitutes a predictable, if not constant, percentage of total body

fat. The amount of subcutaneous fat, i.e., skinfold thickness, has been found to be highly related to the actual body density as measured by underwater weighing techniques (41). Vegetarians have been found to have lower body weights as well as lower skinfold thicknesses than nonvegetarian counterparts (30).

The relationship between body density and skinfold thickness appears to be logarithmic with the regression equation for these 2 parameters depending upon age and sex (42). That is, a given skinfold thickness corresponds to a considerably lower body density in females than in males, implying that in women a lesser proportion of the body fat content is subcutaneous. Decreases in the ratios of subcutaneous fat to total body fat with increasing age (41) and with increasing degree of obesity (42) have been found. Hill et al. (43) reported a very high correlation ($r = 0.91$) between total body nitrogen and fat-free mass as estimated by skinfold thickness. Some investigators prefer to make skinfold thickness measurements at several sites on the body (41). Seltzer and Mayer (44) reported that the triceps skinfold thickness showed the highest correlation with body density measured by underwater weighing. They also point out the desirability of this site of measurement from the aspects of ease of measurement, reproducibility of results and relative lack of inconvenience and embarrassment to the subject.

## VI. URINARY CONSTITUENTS RELATED TO PROTEIN METABOLISM

Urine, the principal excretory route for nitrogenous waste products, contains various metabolites which are derived either from dietary

protein or from endogenous protein metabolism. Measurement of these compounds in urine has been hypothesized as a means of assessing the protein metabolic status of the organism (45). Among these parameters are urea, ammonia, inorganic sulfate, creatinine and hydroxyproline.

Urea

Urinary urea has been found to reflect both the quality and quantity of dietary proteins. Taylor et al. (46) found strong correlations between urea nitrogen excretion and either net protein utilization or dietary protein level. These correlations were strongest when dietary protein was below the maintenance nitrogen level. When protein intake was increased beyond the maintenance nitrogen level, both absolute urea nitrogen and total nitrogen excretion increased, but urea excretion did not necessarily increase when expressed as a percent of the total urinary nitrogen (47). In subjects given the same levels of dietary protein with either sufficient energy intake to maintain body weight or energy intake restricted to 80% of maintenance level, urea excretion was increased with energy restriction.

In animals, urea excretion has been found to increase when rats (48) and pigs (49) were fed diets deficient in 1 or more of the indispensable amino acids. Brown and Cline (49) reported a significant decrease in total urinary urea excretion in pigs fed a diet deficient in either lysine or tryptophan supplemented with that amino acid when compared to pigs fed the same diet without supplementation. Nakagawa and Masana (50) noted increased urinary urea in young men fed energy adequate

isonitrogenous diets deficient in either tryptophan, lysine or methionine. They attributed this increase to an increase in degradation of body proteins even though the creatinine nitrogen excretion remained constant and concluded that the urea nitrogen or the ratio of urea to creatinine may not be used as an index of protein nutrition in slight short-term protein or amino acid deficiency states.

Sulfate

In recognition of the fact that the sulfur amino acids may be the limiting amino acids in the vegetable proteins comprising most of the protein consumed by much of the world's population (51), it has been postulated that the urinary excretion of sulfate may be used as an index of dietary protein quality. It has been suggested that various ratios of urinary sulfate sulfur, urinary nitrogen and/or urinary creatinine may be useful for predicting the nutritive value of dietary proteins (52). Miller and Mumford (53) and Pellett (54) found that 24-hour excretion of total sulfate reflected the protein level of the diet. Lakshmanan et al. (55) reported that urinary inorganic sulfate levels were decreased markedly by feeding a protein-free diet. Bodwell et al. (56) studied the effect of consumption of a constant amount of various proteins on urinary sulfate excretion and concluded that urinary sulfate per se is not a useful index of protein nutritional value in humans at protein intakes greater than 1 g of protein per kg of body weight per day. However, sulfate excretion levels may be useful in a multiple-parameter index for estimating protein nutritive value (51) or in cases of doubtful protein adequacy (52).

Creatinine

Creatinine is the second most abundant nitrogenous compound found in urine. The creatinine excreted appears to be of both dietary and endogenous origin. Crim et al. (57, 58) reported that creatinine excretion decreased when young men were changed from a creatine-containing to a creatine-free diet. The subjects were on a vigorous exercise regimen and exhibited distinctly positive nitrogen balances. The authors concluded that the size of the creatine body pool can be influenced by dietary creatine and that creatinine excretion represents a constant fraction of this pool and can therefore change independently of lean body mass. These data are consistent with those of Bleiler and Schedl (59). Murlin et al. (60) found urinary creatinine excretion was linearly related to the biological value of the protein. Powell et al. (61) noted higher creatinine excretions in subjects on a meat diet compared to subjects on a gluten diet. This difference in creatinine excretion precluded the use of a urinary total nitrogen to creatinine ratio in assessing either protein intake or the biological value of the proteins.

In sheep, Van Niekerk et al. (62) found that urinary creatinine was independent of the type and amount of diet fed, as well as the age, size and amount of body fatness in the animal; that is, creatinine excretion is a constant function of the lean body mass in sheep and hence endogenous protein turnover. Recently, Duggal and Eggum (63) reported that the daily excretion of urinary creatinine in pigs increased almost linearly with body weight, reinforcing the hypothesis that creatinine

excretion is reflective of lean body mass and endogenous protein metabolism (40).

Hydroxyproline

Among the biochemical measurements that have been correlated with endogenous protein metabolism is the urinary excretion of hydroxyproline (HOP). Collagen, the major protein containing this amino acid, is about 13% HOP. It has been hypothesized that, on a minimal hydroxyproline diet such as that consumed by vegetarians, urinary HOP excretion is related to collagen metabolism, particularly that of skeletal collagen (64, 65, 66). Therefore, any normal or pathological condition resulting in changes in collagen metabolism would be reflected by changes in HOP excretion. Saleh and Coengracht (67) reported that in subjects 10 to 20 years of age, HOP excretion was significantly greater than that found in adults. The HOP excretion has been found to be relatively constant from age 21 to 70 and to decrease thereafter independently of skeletal weight (68, 69).

# CHAPTER III

# PROCEDURES

## I. GENERAL PLAN

One hundred twenty-five vegetarian and nonvegetarian females aged 40 years and over were participants in this study. Of the 63 vegetarians, 6 were vegans and 57 were lactoovovegetarians. Some of the women who classified themselves as vegetarians would eat meat occasionally; 4 vegetarians had 1 serving of meat each and 2 had 2 servings of meat each during the week in which they kept dietary records. The 62 nonvegetarians normally consumed 1 or more servings of meat per day; however, 1 had only 4 servings and 2 had 6 servings each of meat during the week in which they recorded their food intakes.

The vegetarian women were recruited primarily from Seventh-Day Adventist churches in Knoxville, Maryville, Collegedale, Greeneville and Wildwood, Tennessee. The nonvegetarians were recruited primarily from Jesus Christ of Latter Day Saints churches in Knoxville and Oak Ridge, Tennessee; from United Methodist churches in Knoxville and Morristown, Tennessee; and from Knox County, Tennessee, Extension Homemakers Clubs.

After having the details of the study explained to her, each subject signed an informed consent form (Appendix A) and completed a dietary history (Appendix A). Each subject completed a 7-day dietary record, collected a 24-hour urine sample and had the following anthropometric measurements made: body height and weight and triceps skinfold

thickness. The project was approved by the Human Rights Committee of The University of Tennessee.

## II. COLLECTION METHODS

### Collection of Dietary Information

Dietary information was obtained from 7-day dietary records (Appendix A) and dietary histories (Appendix A). Each subject was given verbal and written instructions (Appendix A) for measuring and recording dietary intakes. The subjects were supplied with a set of measuring cups, measuring spoons and a plastic ruler to help estimate food portion sizes. The 7-day records were returned in person or by mail to members of the research team.

Food items recorded on the dietary sheets were summarized, coded and the amounts consumed converted to grams. The code numbers used were either those listed in USDA Handbook No. 8 (70) or those that had been established by project workers using food composition values supplied by food manufacturers. Conversion of the food measures such as cups, cubic inches, and so forth, to grams was accomplished using values given in Nutritive Values of American Foods in Common Measures (71), Food Values of Portions Commonly Used (72), as well as data supplied by Loma Linda Foods,[1] Worthington Foods[2] and other food manufacturers. Code numbers and gram weights of the foods consumed were placed on data cards and

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[1]Loma Linda Foods, Loma Linda, CA.

[2]Worthington Foods, Worthington, OH.

total and average daily nutrient intakes were calculated by computer using USDA Handbook No. 8 (70) data and the additional values added by project workers. Nutrient content of food items not occurring on the tape were added manually to the intake of the subject. Nutrient contents of dietary supplements were established using product labels or the Physicians' Desk Reference (73). Values were calculated manually and expressed as a mean for 7 days.

## Urine Collection and Storage

Subjects were given verbal and written instructions (Appendix A) for collecting a 24-hour urine sample. Acid-rinsed (concentrated hydrochloric acid diluted 1:7 with water) polyethylene bottles containing 5 ml of a 10% (w/v) thymol-2-propanol solution as a preservative were supplied. Samples were picked up by or returned to a member of the research team the day of completion of the collection. Samples were kept on ice and returned to the laboratory as soon as possible. Each urine sample was thoroughly mixed, the volume was measured in a 2-liter graduated cylinder, and an aliquot was removed and frozen for analysis.

## Anthropometric Measurements

Height, weight and skinfold thickness were measured on each subject. Height was measured to the nearest 1/4 inch and weight to the nearest pound with the subject dressed in light indoor clothing and wearing no shoes. Height in inches was converted to cm by multiplying by 2.54 and weight in pounds was converted to kg by dividing by 2.2. Skinfold

thickness was measured to the nearest mm using Lange[3] calipers and following the method described by Tanner and Whitehouse (74). Triplicate readings were taken on the left triceps of each subject midway between the tip of the acromion process and the top of the radius. The distance between these 2 points was measured with a steel measuring tape.

Calculation of Percent Body Fat and Lean Body Mass

Percent body fat was calculated according to the formula:

$$Y = 0.934X + 6.16$$

where Y = average triceps skinfold measurement

X = percent body fat (44).

Lean body mass (LBM) was computed according to the formula:

$$LBM = 0.29569W + 0.41813H - 43.2933$$

where W = weight in kg

H = height in cm (39).

## III. CHEMICAL METHODS

Ammonia in Urine

Ammonia nitrogen was measured by the Berthelot reaction in which phenol and alkali-hypochlorite solutions react with the ammonia to produce a blue chromophore whose concentration is then measured spectrophotometrically. The mechanism is unknown but appears to involve the conversion of ammonia to chloramine, with subsequent conjugation and

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[3]Cambridge Scientific Industries, Inc., Cambridge, MD.

oxidation to p-quinonechloramine and indophenol and a reduction to indophenolate ion (75-77). All analyses were done in duplicate.

Reagents. Phenol solution: 60 g A.R. grade phenol ($C_6H_5OH$) were dissolved in demineralized water by heating and 25 mg A.R. grade sodium nitroprusside were added. The solution was cooled and diluted with demineralized water to 1 liter. The solution was stable for about 1 week when stored in the dark.

Alkali-hypochlorite solution: 25 g A.C.S. certified NaOH and 40 ml commercial bleach[4] were combined and diluted with demineralized water to 1 liter.

Ammonia nitrogen standard (0.10 mg $NH_3$ N/ml): 472 mg A.R. grade $(NH_4)_2SO_4$ were dissolved in water and diluted with water to 1 liter.

Procedure. Using micropipets, an 0.025 ml sample of urine was added to a test tube containing 0.5 ml of water. Five ml of phenol solution and 5 ml of alkali-hypochlorite solution were added and the tube contents were mixed. After incubating for 30 minutes at 37°, absorbance of the sample was read at 540 nm against a reagent blank. Standards containing 0, 0.002, 0.005 and 0.010 mg $NH_3$ N/ml were treated similarly.

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[4]Clorox, The Clorox Company, Oakland, CA.

Calculations.

$$\text{mg } NH_3 \text{ N/ml of urine} = \frac{\text{Absorbance of sample}}{\Sigma \text{ absorbance of standards}} \times \Sigma \text{ concentration of standards (mg/ml)}$$

$$\times \text{ (dilution factor = 40)}$$

$$\text{mg } NH_3 \text{ N/day} = \text{mg } NH_3 \text{ N/ml} \times \text{24-hour urine volume (ml)}.$$

Creatinine in Urine

Urinary creatinine was measured using a modification of Folin's method (78). This method is based on the Jaffe reaction in which creatinine reacts with an alkaline picrate solution to form a tautomer of creatinine picrate. The concentration of the red chromophore is then measured spectrophotometrically. All analyses were done in duplicate.

Reagents. Picric acid (0.04 M): 10.17 g A.R. grade picric acid ($(NO_2)_3C_6H_2OH$) were dissolved in water and the volume brought to 1 liter with water.

Sodium hydroxide (0.75 N): 30 g A.C.S. certified NaOH were dissolved in water and the volume brought to 1 liter with water.

Hydrochloric acid (0.1 N): 8.3 ml A.C.S. reagent concentrated HCl (approximately 12 N) were diluted with water to 1 liter.

Alkaline-picrate solution: 0.04 M picric acid and 0.75 N NaOH were mixed 1:1 just prior to use.

Stock creatinine standard (1500 µg/ml): 150 mg creatinine hydrochloride ($CH_3NCNHNHCOCH_2 \cdot HCl$) were dissolved in 100 ml of 0.1 N HCl.

Working creatinine standard (15 µg/ml): 1 ml stock creatinine standard was diluted to 100 ml with water. This solution was prepared fresh daily.

Procedure. A 1 ml sample of urine was diluted to 100 ml with water. A 3 ml aliquot was removed and mixed with 2 ml alkaline-picrate solution. After 20 minutes, absorbance was read at 520 nm against a reagent blank. Standards containing 0, 7.5, 15.0, 30.0 and 45.0 µg/tube were treated similarly.

Calculations.

$$\mu g \text{ creatinine/ml of urine} = \frac{\text{Absorbance of sample}}{\Sigma \text{ absorbance of standards}} \times \begin{matrix}\Sigma \text{ concentration} \\ \text{of standards} \\ (\mu g/ml)\end{matrix}$$

$$\times \text{ (dilution factor = 33.3)}$$

$$\text{g creatinine/day} = \frac{\mu g \text{ creatinine/ml}}{10^6} \quad \text{24-hour urine volume (ml).}$$

Total Hydroxyproline in Urine

Urinary total hydroxyproline was determined using a modification of the method described by Bergman and Loxley (79). Acid-hydrolyzed urine samples were neutralized, the hydroxyproline was oxidized to pyrrole with chloramine T and reacted with Ehrlich's reagent (p-dimethyl-aminobenzaldehyde). The concentration of the resulting red chromophore was then measured spectrophotometrically. All determinations were done in duplicate.

Reagents. Hydroxyproline standard solution (150 mg hydroxyproline/liter): 150 mg L (-) hydroxyproline[5] were dissolved in water and the volume brought to 1 liter with water.

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[5]Eastman Chemicals Corp., Rochester, N.Y.

Hydrochloric acid: A.C.S. certified concentrated HCl (approximately 12 N).

Phenolphthalein (1% w/v): 1 g A.R. grade phenolphthalein was dissolved in 95% ethanol and the volume brought to 100 ml.

Saturated lithium hydroxide (w/w): 127 g anhydrous LiOH were dissolved in 1 liter of water.

n-Propanol: A.R. grade.

Citrate-acetate buffer: 57 g A.C.S. certified sodium acetate trihydrate ($NaC_2H_3O_2 \cdot 3H_2O$), 37.5 g A.C.S. certified trisodium citrate dihydrate ($Na_3C_6H_5O_7 \cdot 2H_2O$), 5.5 g A.C.S. certified citric acid monohydrate ($C_6H_8O_7 \cdot H_2O$) and 385 ml A.R. grade n-propanol were dissolved in water and the volume brought to 1 liter with water. This solution was stable indefinitely.

Aqueous chloramine T (7% w/v): 7 g chloramine T[6] ($C_7H_7ClNO_2SNa \cdot 3H_2O$) were dissolved in water and the volume brought to 100 ml with water. This solution was prepared fresh daily.

Oxidant solution: Aqueous chloramine T and citrate-acetate buffer were mixed 1:4 just prior to use.

Ehrlich's reagent: 17.6 g certified p-dimethylaminobenzaldehyde[7] ($C_9H_{11}OH$) were dissolved in 40.8 g A.C.S. reagent perchloric acid (60% $HClO_4$, S.G. 1.54) and the volume brought to 100 ml with A.R. grade n-propanol immediately prior to use.

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[6]Fisher Scientific Company, Norcross, GA.

[7]Fisher Scientific Company, Norcross, GA.

Procedure. Five ml of urine were placed in each of 4 15-ml screw-capped borosilicate tubes. One ml water was added to 2 of the tubes; 1 ml hydroxyproline standard solution was added as an internal standard to the other 2 tubes. Five ml concentrated HCl were added to each tube, the tubes were tightly capped, placed in sand baths and heated overnight in an oven at 105 ± 5°. After cooling, the hydrolysate was transferred to a 25-ml volumetric flask and 3 to 5 drops of 1% phenolphthalein added. The sample was then neutralized to the phenolphthalein end point with saturated LiOH, made slightly acidic again with 6 N HCl and the volume brought to 25 ml with water. The neutralized hydrolysate was then filtered through a sintered glass funnel. One ml of the filtered hydrolysate was placed in a test tube containing 2 ml n-propanol. While mixing, 1 ml oxidant solution was added and the sample was incubated for 4.0 ± 0.5 minutes at room temperature before the addition of 2 ml Ehrlich's reagent. The sample was thoroughly mixed and placed in a 60° water bath for 21 minutes. After an hour at room temperature, absorbance was read at 562 nm against a reagent blank.

Calculations.

$$\text{mg hydroxyproline/ml of urine} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard - absorbance of sample}} \times \text{concentration of standard (mg/ml)} \times (\text{dilution factor} = 33.3).$$

$$\text{mg hydroxyproline/day} = \text{mg hydroxyproline/ml} \times \text{24-hour urine volume (ml)}.$$

Nitrogen in Urine

Urinary nitrogen was measured using the macro-Kjeldahl method (80). Nitrogenous compounds in the urine were converted to ammonium sulfate; ammonia was liberated by distilling in the presence of sodium hydroxide, collected in boric acid and titrated with standardized hydrochloric acid. All analyses were done in duplicate.

Reagents. Sodium sulfate: Anhydrous A.C.S. certified $Na_2SO_4$.

Cupric sulfate: A.C.S. certified $CuSO_4 \cdot 5H_2O$.

Concentrated sulfuric acid: A.C.S. certified $H_2SO_4$ (approximately 36N).

Sodium hydroxide (50% w/v): 500 g A.C.S. certified NaOH were dissolved in 750 ml water, allowed to cool and the volume brought to 1 liter with water.

Saturated boric acid: 55 g A.C.S. certified boric acid ($H_3BO_3$) were dissolved, with heating, in water and the volume brought to 1 liter with water.

Methyl red-methylene blue indicator: 2 parts of 0.2% alcoholic methyl red were combined with 1 part of 0.2% alcoholic methylene blue.

Hydrochloric acid (0.1 N): 8.3 ml of A.C.S. certified concentrated HCl (approximately 12 N) were diluted to 1 liter with water and standardized (81).

Procedure. A 5 ml sample of urine was added to a Kjeldahl flask containing 5 g sodium sulfate, approximately 0.3 g cupric sulfate, a

Hengar crystal[8] and 2 glass beads. Sixteen ml of concentrated sulfuric acid were layered down the side of the flask. Flask contents were digested until the solution was clear (approximately 2 hours). The digest was cooled to room temperature, 200 ml of water were added, the sample was well mixed and it was again cooled. Seventy-five ml of 50% sodium hydroxide were poured down the side of the flask so as to minimize mixing with the digest. The flask was connected to a distillation rack, contents were swirled to mix, the burner was ignited and the ammonia was distilled into an Erlenmeyer flask containing 50 ml saturated boric acid and 3 drops of methyl red-methylene blue indicator. Distillation was continued until the Erlenmeyer flask contained approximately 125 ml. The ammonium borate in the collecting flask was titrated with standardized 0.1 N hydrochloric acid to the color of a simultaneously run blank.

Calculations. $$\text{mg N/ml of urine} = \frac{\text{ml HCl used} \times \text{N of HCl} \times 14}{\text{ml of urine used}}$$

$$\text{g N/day} = \frac{\text{mg N/ml}}{10^3} \times \text{24-hour urine volume (ml)}.$$

Inorganic Sulfate in Urine

Inorganic sulfate was measured in a diluted urine sample by the addition of barium and sodium rhodizonate. The rhodizonate reacts with barium ions to give an orange-red colored complex, the intensity of which may be measured spectrophotometrically. The intensity of this color is proportional to the concentration of barium ions. In the presence of

[8]Hengar Company, Philadelphia, PA.

inorganic sulfate, barium ions are complexed to the sulfate and the intensity of the color is lowered in a direct relationship to the amount of barium sulfate formed (82). All analyses were done in duplicate.

Reagents. Sulfate standard solution (30 µg sulfate/ml): 44.4 mg A.R. grade $Na_2SO_4$ were dissolved in demineralized water and the volume brought to 1 liter with demineralized water.

Acetic acid (2 M): 114 ml A.C.S. reagent glacial acetic acid (approximately 18 N) were diluted to 1 liter with water.

Barium chloride (0.005 M): 1.04 g A.R. grade $BaCl_2$ were dissolved in water and the volume brought to 1 liter with water.

Sodium bicarbonate (0.02 M): 1.680 g A.R. grade $NaHCO_3$ were dissolved in water and the volume brought to 1 liter with water.

Barium chloride solution: One hundred ml 2 M $CH_3COOH$, 40 ml 0.005 M $BaCl_2$ and 80 ml 0.02 M $NaHCO_3$ were mixed and the volume brought to 1 liter with absolute ethanol.

Sodium rhodizonate solution: 5 mg disodium rhodizonate[9] were dissolved in 20 ml water, 100 mg L-ascorbic acid were added and the solution was thoroughly mixed. The volume was made up to 100 ml with absolute ethanol. The solution was used promptly after being allowed to set for 30 minutes.

Procedure. All glassware was washed in nitric acid (concentrated nitric acid diluted 1:4 with water). A 1 ml sample of urine was diluted

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[9]J. T. Baker Chemical Company, Phillipsburg, N.J.

to 100 ml with water. An 0.5 ml aliquot of the diluted urine was added to 2.0 ml absolute ethanol. One ml of barium chloride solution was added and the sample was allowed to stand for at least 5 minutes. A 1.5 ml sample of sodium rhodizonate solution was added, tube contents were mixed and allowed to stand at room temperature in the dark for 10 minutes. The absorbances of the samples and the standards were read at 520 nm against water. Samples were read as soon as possible after 10 minutes since the color began to fade after 30 minutes. Standards containing 0, 3.0, 6.0, 12.0 and 15.0 μg $SO_4$/ml were treated similarly.

Calculations.

μg $SO_4$/ml of urine = B(absorbance of blank - absorbance of sample) + A × (dilution factor = 200)

where B = slope of the line for (absorbance of blank - absorbance of standard) plotted vs. concentration of the standards

A = intercept of this line.

$$\mu g\ SO_4/day = \frac{\mu g\ SO_4/ml}{10^6} \times \text{24-hour urine volume (ml)}.$$

Urea Nitrogen in Urine

Urea in a diluted urine sample was converted to ammonia and carbon dioxide by urease. Ammonia nitrogen was then measured by the Berthelot reaction (see Ammonia in Urine determination, p. 18) (75-77). True urea nitrogen was obtained by correcting for urinary ammonia nitrogen. All analyses were done in duplicate.

Reagents. EDTA buffer, pH 6.5 (27 mM): 1 g ethylenediamine tetraacetic acid disodium salt ($C_{10}H_{18}N_2O_{10}Na_2$)[10] was dissolved in 90 ml demineralized water. The pH was adjusted to 6.5 with NaOH and the volume diluted with water to 100 ml.

Urease solution: 20 mg urease[11] were dissolved in EDTA buffer and diluted to 50 ml. The solution was kept at 4° and prepared fresh every 2 days.

Urea nitrogen standard (20 mg urea N/100 ml): 428 mg A.R. grade urea[12] were dissolved in and diluted with a cold saturated solution of benzoic acid in demineralized water to 1 liter.

For other reagents, see Ammonia in Urine determination, p. 18.

Procedure. A 5 ml sample of urine was diluted to 100 ml. Using micropipets, 0.025 ml of diluted urine was added to a test tube containing 0.5 ml urease solution. The tube was incubated in a 37° water bath for 30 minutes. Five ml of phenol solution and 5 ml of alkali hypochlorite solution were added and the tube contents were mixed. After 30 minutes at room temperature, absorbance of the sample was read against a reagent blank at 590 nm. Standards containing 0, 0.2, 0.5, 1.0 and 2.0 µg urea N/ml were treated similarly.

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[10]Fisher Scientific Company, Norcross, GA.

[11]Soluble urease, 1500 Sumner units/g. ICN Pharmaceuticals, Inc., Life Sciences Group, Cleveland, OH.

[12]Mallinckrodt Chemical Works, St. Louis, MO.

Calculations.

$$\mu g \text{ apparent urea N/ml of urine} = \frac{\text{Absorbance of sample}}{\Sigma \text{ absorbance of standards}} \times \Sigma \text{ concentration of standards } (\mu g/ml) \times (\text{dilution factor} = 800)$$

$$\mu g \text{ true urea N/ml of urine} = \mu g \text{ apparent urea N/ml} - \mu g \text{ ammonia N/ml}$$

$$g \text{ urea N/day} = \frac{\mu g \text{ true urea N/ml}}{10^6} \times \text{24-hour urine volume (ml)}.$$

## IV. STATISTICAL ANALYSES

The Statistical Analyses System (SAS) (83), a computer software package, was used to perform statistical analyses. The data were adjusted to the mean age or the mean daily protein intake of the sample using a least squares procedure. This procedure derives a set of values for the data that minimizes the sums of squares between the observed value and the predicted value. Tests for significance of differences among mean values were done using Duncan's New Multiple Range Test (84). Correlation coefficients reported are Pearson r values determined by computer. In determining the regression model which best described the relationship between age and each of the other variables, a backward elimination technique was employed. The linear, quadratic and cubic functions of age and the interactions of these functions with classification as vegetarians or nonvegetarians constituted the original model. The probability level of the partial F statistic based on Type I (sequential) sums of squares was determined for each term in the model.

The partial F statistic and the Type I sum of squares assessed the additional contribution of that variable to the prediction of the dependent variable over and above what is contributed by the other variables in the given model. The highest level term producing a significant ($P < 0.05$) F statistic became the final term for that model. A regression equation was then recalculated using this revised model and was considered to be the best estimate of the relationship between age and that variable.

# CHAPTER IV

## RESULTS

There were 125 subjects in this study of the relationships between age, the consumption of a vegetarian diet, physical measurements and the urinary excretion of protein-derived metabolites. Of these subjects, 63 were vegetarian and 62 were nonvegetarian. The vegetarian group included 57 lactoovovegetarians and 6 vegans. Although most of the women who classified themselves as vegetarians consumed no meat, during the week in which they kept their dietary records 4 of the lactoovovegetarians had 1 serving of meat each and 2 had 2 servings each. Most of the 62 nonvegetarian subjects consumed at least 1 serving of meat per day; however, during the week in which they kept their dietary records, 1 had only 4 servings of meat and 2 had 6 servings of meat each. The vegetarian women had followed that dietary regimen for at least 2 years, with a mean duration of vegetarianism of 36.4 years. Throughout the remainder of the text, the following abbreviations will be used to indicate the groups of subjects: V—vegan subjects (n = 6), LOV—lactoovovegetarian subjects (n = 57), AV—all vegetarian subjects (LOV + V, n = 63) and NV—nonvegetarian subjects (n = 62).

Physical measurements adjusted to the mean age of the sample (59.4 years) are given in Table I. The mean heights (± SEM) of the groups NV, V and LOV were 161.1 ± 0.8, 162.8 ± 2.4 and 162.0 ± 0.8 cm, respectively. These values were not significantly different. The V group (51.0 ± 5.6 kg) weighed significantly less than the LOV

TABLE I

PHYSICAL MEASUREMENTS[1] OF OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS ADJUSTED TO THE MEAN AGE[2] OF THE SAMPLE

| Parameter | Vegetarians | | | Nonvegetarians[3] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[4] (n = 57) | (n = 62) |
| Height (cm) | 161.5 ± 0.8[a5] | 162.8 ± 2.4[a] | 162.0 ± 0.8[a] | 161.1 ± 0.8[a] |
| Weight (kg) | 62.7 ± 1.5[a] | 51.0 ± 5.6[b] | 65.8 ± 1.8[a] | 66.7 ± 1.6[a] |
| Triceps Skinfold Thickness (mm) | 26.8 ± 1.0[a] | 19.8 ± 3.6[b] | 28.5 ± 1.1[a] | 32.4 ± 1.0[c] |
| Body Fat (%)[6] | 22.3 ± 1.0[a] | 14.8 ± 3.9[b] | 24.1 ± 1.2[a] | 28.1 ± 1.1[c] |
| Lean Body Mass (kg)[7] | 42.8 ± 0.6[a] | 39.9 ± 2.1[b] | 43.9 ± 0.7[a] | 43.7 ± 0.6[a] |

[1]Mean ± SEM.

[2]59.4 years.

[3]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[4]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[5]Means in a row not sharing a common superscript are significantly different ($P < 0.05$)

[6]Calculated using the formula: triceps skinfold thickness = 0.934 (percent body fat) + 6.16.

[7]Calculated using the formula: LBM = 0.29569 (weight) + 0.41813 (height) - 43.2933.

(65.8 ± 1.8 kg) and NV (66.7 ± 1.6 kg); the mean weights of the latter 2 groups did not differ. Triceps skinfold thickness for V was 19.8 ± 3.6 mm, which was significantly less than the 28.5 ± 1.1 mm value for the LOV. The skinfold thickness of NV (32.4 ± 1.0 mm) was significantly larger than that of the vegetarian groups. The percent body fat of the V (14.8 ± 3.9%) was significantly smaller than LOV (24.1 ± 1.2%), while that of NV (28.1 ± 1.1%) was significantly larger than that of either group of vegetarians. The lean body mass (LBM) of V (39.9 ± 2.1 kg) was also significantly less than those of LOV (43.9 ± 0.7 kg) and NV (43.7 ± 0.6 kg). The LBM of the NV group did not differ significantly from that of LOV.

Table II presents age-adjusted mean daily energy and protein intakes for the subjects. The V, LOV and NV groups had statistically similar energy intakes of 1601 ± 133 kcal/day, 1509 ± 42 kcal/day and 1524 ± 42 kcal/day, respectively. The total daily protein intake of the V group was 52.2 ± 4.2 g and for LOV was 54.6 ± 1.3 g; these values were not significantly different. The NV (66.5 ± 1.6 g/day) consumed significantly more protein than the vegetarian groups. The LOV group (14.7 ± 0.3%) derived a significantly greater percentage of their total energy from protein than the V (13.0 ± 0.9%); the 17.8 ± 0.3% value for the NV was significantly greater than that of the vegetarian groups. Animal protein intake for the LOV group was significantly less than that of NV when it was expressed either in absolute amounts or as a percentage of total protein intake. The LOV consumed 16.0 ± 1.7 g or 29.5 ± 2.1% of total protein from animal sources and NV consumed

TABLE II

MEAN DAILY ENERGY AND PROTEIN INTAKES[1] OF OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS ADJUSTED TO THE MEAN AGE[2] OF THE SAMPLE

| Nutrient | Vegetarians | | | Nonvegetarians[3] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[4] (n = 57) | (n = 62) |
| Energy (kcal/day) | 1500 ± 42[a5] | 1601 ± 133[a] | 1509 ± 42[a] | 1524 ± 42[a] |
| Total Protein (g/day) | 53.6 ± 1.6[a] | 52.2 ± 4.2[a] | 54.6 ± 1.3[a] | 66.5 ± 1.6[b] |
| Animal Protein (g/day) | 14.8 ± 1.6[a] | 4.4 ± 5.2[b6] | 16.0 ± 1.7[a] | 45.8 ± 1.7[c] |
| Vegetable Protein (g/day) | 29.1 ± 1.1[a] | 38.2 ± 3.2[b] | 28.2 ± 1.1[a] | 10.8 ± 1.1[c] |
| Mixed Protein (g/day)[7] | 10.5 ± 0.6[a] | 9.9 ± 1.9[a] | 10.5 ± 0.6[a] | 10.3 ± 0.6[a] |
| Energy from Protein | 14.5 ± 0.3[a] | 13.0 ± 0.9[b] | 14.7 ± 0.3[a] | 17.8 ± 0.3[c] |
| Protein Sources: | | | | |
| Animal (% of total) | 27.2 ± 2.1[a] | 6.3 ± 6.4[b6] | 29.5 ± 2.1[a] | 68.2 ± 2.2[c] |
| Vegetable (% of total) | 54.1 ± 1.7[a] | 74.8 ± 5.0[b] | 52.1 ± 1.7[a] | 16.4 ± 1.8[c] |
| Mixed (% of total) | 19.8 ± 1.0[a] | 19.5 ± 3.1[a] | 19.6 ± 1.0[a] | 15.6 ± 1.0[b] |

[1]Mean ± SEM.

[2]59.4 years.

[3]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[4]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[5]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

[6]Appearance of consumption of animal protein by the vegans is due to calculation errors.

[7]Includes foods containing both animal and vegetable proteins.

45.8 ± 1.7 g or 68.2 ± 2.2%. The appearance in the tables of animal and mixed protein intakes for the subjects classified as vegans is a result of coding food intakes using USDA Handbook No. 8 (70) numbers for some mixed dishes in an attempt to obtain the best possible overall nutrient intake data. These figures, therefore, represent an artifact of the analysis rather than the actual intakes of the subjects. Vegetable protein intake was significantly greater for V (38.2 ± 3.2 g/day or 74.8 ± 5.0% of total protein) than for LOV (28.2 ± 1.1 g or 52.1 ± 1.7%). The NV consumed 10.8 ± 1.1 g or 16.4 ± 1.8% of total protein from vegetable sources which was significantly lower than the intakes of the vegetarian groups.

Table III presents age-adjusted mean daily excretions of urinary components. Mean daily total nitrogen excretion for the V, LOV and NV groups were 8.50 ± 1.10, 8.49 ± 0.35 and 9.00 ± 0.36 g of total nitrogen/day; these values were statistically similar. Urea nitrogen excretion did not differ among the groups when expressed either as an absolute amount or as a percentage of the total nitrogen. The NV excreted 7.77 ± 0.36 g or 86.5 ± 1.6% of total nitrogen as urea nitrogen; the V, 7.08 ± 1.09 g or 83.9 ± 5.1%; and the LOV, 7.24 ± 0.36 g or 85.5 ± 1.7%, respectively. The ammonia nitrogen excretion of the LOV (174.6 ± 13.4 mg/day) was significantly less than that of the NV (226.6 ± 15.1 mg), while ammonia nitrogen excretion of the V (201.2 ± 42.0 mg) did not differ from that of either group. The LOV (2.11 ± 0.15%) excreted less of the total nitrogen as ammonia than either NV (2.64 ± 0.16%) or V (2.59 ± 0.48%). The latter values (for V and NV) did not significantly

TABLE III

MEAN DAILY EXCRETION[1] OF URINARY COMPONENTS BY OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS ADJUSTED TO THE MEAN AGE[2] OF THE SAMPLE

| Metabolite | Vegetarians | | | Nonvegetarians[3] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[4] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 8.20 ± 0.36[a5] | 8.50 ± 1.10[a] | 8.49 ± 0.35[a] | 9.00 ± 0.36[a] |
| Urea Nitrogen (g/day) | 6.92 ± 0.36[a] | 7.08 ± 1.09[a] | 7.24 ± 0.36[a] | 7.77 ± 0.36[a] |
| Ammonia Nitrogen (mg/day) | 170.3 ± 15.1[a] | 201.2 ± 42.0[ab] | 174.6 ± 13.4[a] | 226.6 ± 15.1[b] |
| Creatinine (g/day) | 1.31 ± 0.05[a] | 1.30 ± 0.14[a] | 1.33 ± 0.04[a] | 1.54 ± 0.05[b] |
| Hydroxyproline (mg/day) | 32.1 ± 1.7[ab] | 28.3 ± 4.8[a] | 33.2 ± 1.6[ab] | 35.5 ± 1.7[b] |
| Inorganic Sulfate (g/day) | 1.49 ± 0.07[a] | 1.41 ± 0.22[a] | 1.55 ± 0.07[a] | 1.57 ± 0.07[a] |
| Nitrogen Sources | | | | |
| Urea (% of total) | 84.0 ± 1.6[a] | 83.9 ± 5.1[a] | 85.5 ± 1.7[a] | 86.5 ± 1.6[a] |
| Ammonia (% of total) | 2.12 ± 0.16[a] | 2.59 ± 0.48[b] | 2.11 ± 0.15[a] | 2.64 ± 0.16[b] |
| Creatinine (% of total) | 6.48 ± 0.32[ab] | 5.96 ± 1.01[a] | 6.39 ± 0.22[ab] | 6.99 ± 0.32[b] |

[1]Mean ± SEM.

[2]59.4 years.

[3]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[4]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[5]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

differ from each other. The V excreted 1.30 ± 0.14 g of creatinine/day and the LOV excreted 1.33 ± 0.04 g/day; these figures did not differ significantly from each other but were significantly less than the 1.54 ± 0.05 g excreted by NV. The V group excreted 5.96 ± 1.01% of total nitrogen as creatinine which did not differ significantly from LOV (6.39 ± 0.22%) but was significantly less than that of the NV (6.99 ± 0.32%). The values for LOV and NV did not differ significantly from each other. The same relationship was found with hydroxyproline (HOP) excretion data, i.e., the V excreted 28.3 ± 4.8 mg of HOP/day, which was statistically similar to LOV (33.2 ± 1.6 mg) but significantly less than NV (35.5 ± 1.7 mg). HOP excretion by the LOV and NV groups did not differ significantly. Inorganic sulfate excretion was statistically similar with the groups V, LOV and NV excreting 1.41 ± 0.22, 1.55 ± 0.07 and 1.57 ± 0.07 g of inorganic sulfate/day, respectively.

Table IV presents mean daily urinary excretions by the groups adjusted to the mean protein intake (60.0 g) of the sample. Total nitrogen excretion for the groups was statistically similar with the V excreting 8.65 ± 1.01 g; the LOV, 8.50 ± 0.33 g; and the NV, 8.59 ± 0.36 g. Urea nitrogen excretion did not differ significantly among the groups when expressed either as an absolute amount or as a percentage of the total nitrogen. The values were 7.25 ± 0.34 g for LOV, 7.26 ± 1.02 g for V and 7.41 ± 0.37 g for NV. Corresponding values for the percentage of total nitrogen as urea were 85.5 ± 1.6%, 83.8 ± 5.0%, and 85.8 ± 1.6%, respectively. Ammonia nitrogen excretion for the LOV group was 175.0 ± 13.1 mg (2.12 ± 0.15% of total nitrogen) which was significantly less

TABLE IV

MEAN DAILY EXCRETION OF URINARY COMPONENTS[1] BY OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS ADJUSTED TO THE MEAN DAILY PROTEIN INTAKE[2] OF THE SAMPLE

| | Vegetarians | | | Nonvegetarians[3] |
|---|---|---|---|---|
| Metabolite | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[4] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 8.60 ± 0.36[a5] | 8.65 ± 1.01[a] | 8.50 ± 0.33[a] | 8.59 ± 0.36[a] |
| Urea Nitrogen (g/day) | 7.30 ± 0.36[a] | 7.26 ± 1.02[a] | 7.25 ± 0.34[a] | 7.41 ± 0.37[a] |
| Ammonia Nitrogen (mg/day) | 170.5 ± 15.7[a] | 197.8 ± 40.7[ab] | 175.0 ± 13.1[a] | 226.5 ± 15.8[b] |
| Creatinine (g/day) | 1.37 ± 0.05[a] | 1.24 ± 0.14[a] | 1.34 ± 0.04[a] | 1.49 ± 0.05[b] |
| Hydroxyproline (mg/day) | 33.0 ± 1.8[ab] | 29.1 ± 4.5[a] | 33.1 ± 1.5[ab] | 34.6 ± 1.7[b] |
| Inorganic Sulfate (g/day) | 1.58 ± 0.07[a] | 1.39 ± 0.22[a] | 1.55 ± 0.07[a] | 1.48 ± 0.08[a] |
| Nitrogen Sources | | | | |
| Urea (% of total) | 84.7 ± 1.6[a] | 83.8 ± 5.0[a] | 85.5 ± 1.6[a] | 85.8 ± 1.6[a] |
| Ammonia (% of total) | 2.03 ± 0.16[a] | 2.45 ± 0.47[ab] | 2.12 ± 0.15[a] | 2.73 ± 0.16[b] |
| Creatinine (% of total) | 6.52 ± 0.33[a] | 5.50 ± 1.00[b] | 6.42 ± 0.32[a] | 6.92 ± 0.33[a] |

[1]Mean ± SEM.

[2]60.0 g.

[3]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[4]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[5]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

than that of NV (226.5 ± 15.8 mg and 2.73 ± 0.16%). The V excreted 197.8 ± 40.7 mg (2.45 ± 0.47% of total nitrogen); the figures for the V group did not differ significantly from that of either of the other groups. When expressed as an absolute amount, creatinine excretion for the vegetarian groups was significantly less than that of the NV. The figures were V, 1.24 ± 0.14 g; LOV, 1.34 ± 0.04 g; and NV, 1.49 ± 0.05 g. When creatinine was expressed as a percentage of total nitrogen, the LOV (6.42 ± 0.32%) and the NV (6.92 ± 0.33%) were statistically similar and the V (5.50 ± 1.00%) excreted significantly less of their total nitrogen as creatinine. HOP excretion by the V was 29.1 ± 4.5 mg which was not significantly different from that of the LOV (33.1 ± 1.5 mg) but was significantly less than that of the NV (34.6 ± 1.7 mg). The difference in HOP excretion by LOV and NV groups was not significant. Inorganic sulfate excretion was 1.39 ± 0.22 g for V, 1.55 ± 0.07 g for LOV and 1.48 ± 0.08 g for NV. These values were not significantly different.

The unadjusted data for the physical measurements, dietary intakes and urinary excretions for the subjects are presented in Appendix B.

Table V presents mean values for ratios of urinary components considered to be related to protein quality. The values in Table V are adjusted to the mean age of the sample. Unadjusted data are given in Appendix B. The inorganic sulfate/total nitrogen (S/N) ratios for the groups were not significantly different from each other with mean values for the V, 0.16 ± 0.02; LOV, 0.18 ± 0.01; and NV, 0.18 ± 0.01. The V group (0.19 ± 0.03) had a significantly lower inorganic sulfate/urea

TABLE V

RATIOS OF URINARY METABOLITES[1] IN OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS ADJUSTED TO THE MEAN AGE[2] OF THE SAMPLE

| Ratio | Vegetarians | | | Nonvegetarians[3] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[4] (n = 57) | (n = 62) |
| Inorganic Sulfate/ Total Nitrogen | 0.18 ± 0.01[a5] | 0.16 ± 0.02[2] | 0.18 ± 0.01[a] | 0.18 ± 0.01[a] |
| Inorganic Sulfate/ Urea Nitrogen | 0.22 ± 0.01[a] | 0.19 ± 0.03[b] | 0.22 ± 0.01[a] | 0.21 ± 0.01[a] |
| Inorganic Sulfate/ Creatinine | 1.17 ± 0.05[a] | 1.12 ± 0.16[a] | 1.20 ± 0.06[a] | 1.05 ± 0.05[a] |

[1]Mean ± SEM.

[2]59.4 years.

[3]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[4]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[5]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

nitrogen (S/U) ratio than LOV (0.22 ± 0.01) and NV (0.21 ± 0.01). The values for LOV and NV did not differ significantly. Inorganic sulfate/creatinine (S/C) ratios did not differ significantly with values for the groups V, LOV and NV of 1.12 ± 0.16, 1.20 ± 0.06 and 1.05 ± 0.05, respectively.

Appendix C contains correlation coefficients for protein intake and the urinary excretion of various metabolites and the ratios discussed above. Total nitrogen excretion was found to be significantly positively correlated with protein intake in both vegetarian groups as was urinary urea nitrogen in the LOV. HOP excretion was also significantly positively correlated with protein intake in the LOV as was urinary inorganic sulfate in the V and NV. Among the ratios examined, the only significant correlation coefficients were between protein intake and the S/N ratio which was positively correlated in the NV. Significant positive correlation coefficients were found between animal protein intake and urinary total nitrogen and urea nitrogen in the NV, between inorganic sulfate and animal protein intake in the LOV and NV and between HOP excretion and animal protein intake in the LOV. Among the ratios studied, the S/N and S/U ratios were significantly positively correlated with animal protein intake in the LOV. The only significant correlations between the level of vegetable protein intake and urinary excretion of the metabolites and ratios studied was with the S/C ratio in V.

Table VI gives the analysis of variance (ANOVA) used in developing the regression models that best described the relationship between age and the physical measurements studied. Both AV and NV groups decreased

TABLE VI

ANALYSIS OF VARIANCE OF PHYSICAL MEASUREMENTS IN OLDER ADULT FEMALE VEGETARIANS AND NONVEGETARIANS

| Variable | df | Mean Squares | | | | |
|---|---|---|---|---|---|---|
| | | Height (cm) | Weight (kg) | Skinfold Thickness (mm) | Body Fat (%) | Lean Body Mass (kg) |
| Type[1] | 1 | 41.9 | 248.6 | 701.8*** | 743.0*** | 3.0 |
| Age | 1 | 440.0*** | 921.9** | 423.2** | 539.5** | 308.7*** |
| $Age^2$ | 1 | | 477.9 | 279.7* | 287.7* | 44.2 |
| $Age^3$ | 1 | | | | | |
| Age*Type | 1 | | 94.6 | 142.8 | 137.4 | 5.6 |
| $Age^2$*Type | 1 | | 1185.3** | 263.8* | 284.1* | 187.0** |
| $Age^3$*Type | 1 | | | | | |
| Residual | | 39.9 | 132.2 | 53.8 | 62.9 | 21.4 |
| $R^2$ | | 0.09 | 0.16 | 0.22 | 0.21 | 0.18 |

[1]Denotes vegetarian and nonvegetarian diet effects.

*P < 0.05.

**P < 0.01.

***P < 0.001.

in height linearly with age (Figure 1); the rates of loss between the 2 groups were not significantly different. Weight was also significantly related to the linear function of age and to the interaction term between the quadratic function of age and type of diet. Figure 2 shows the regression lines for the AV and NV groups. The NV tended to have a lower weight with increasing age until about age 70 when a tendency toward increasing weight was observed. The reverse was true in the AV who tended to have the greatest body weight at age 60 years and then decline. There was a significant difference between AV and NV groups in skinfold thickness which was related to age, the quadratic function of age and the interaction term between age and type of diet (Figure 3). The NV showed a gradual decrease in skinfold thickness throughout the age span studied. The AV had a peak skinfold thickness about age 60 followed by a gradual loss. The regression of body fat on age followed the same trend as that of skinfold thickness on age. Lean body mass showed highly significant relationships with age and with the interaction term between the quadratic function of age and type of diet. The plot of this relationship showed that the NV reached a minimum LBM between ages 60 and 70 followed by a slight increase. The reverse pattern was followed by the AV (Figure 4).

Table VII presents the ANOVA used in developing the regression equations for dietary intakes of protein and energy on age. There was no significant difference in mean energy intakes between the AV and NV. There was a significant relationship with the linear function of age and with the interaction term for the cubic function of age and type of diet.

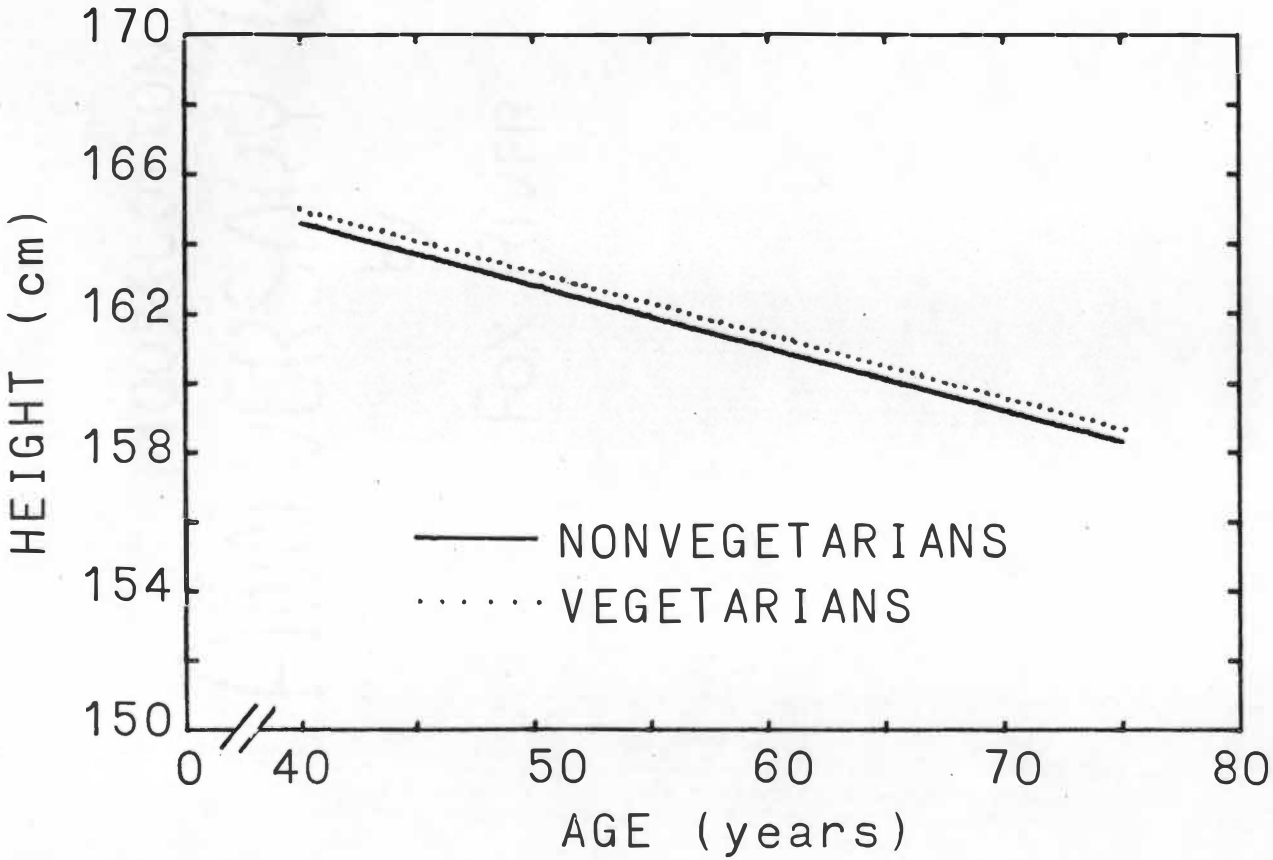


Figure 1. Regression of height on age in adult female vegetarians and nonvegetarians. Height: nonvegetarians = 171.72 - 0.18 (age); vegetarians = 172.17 - 0.18 (age).

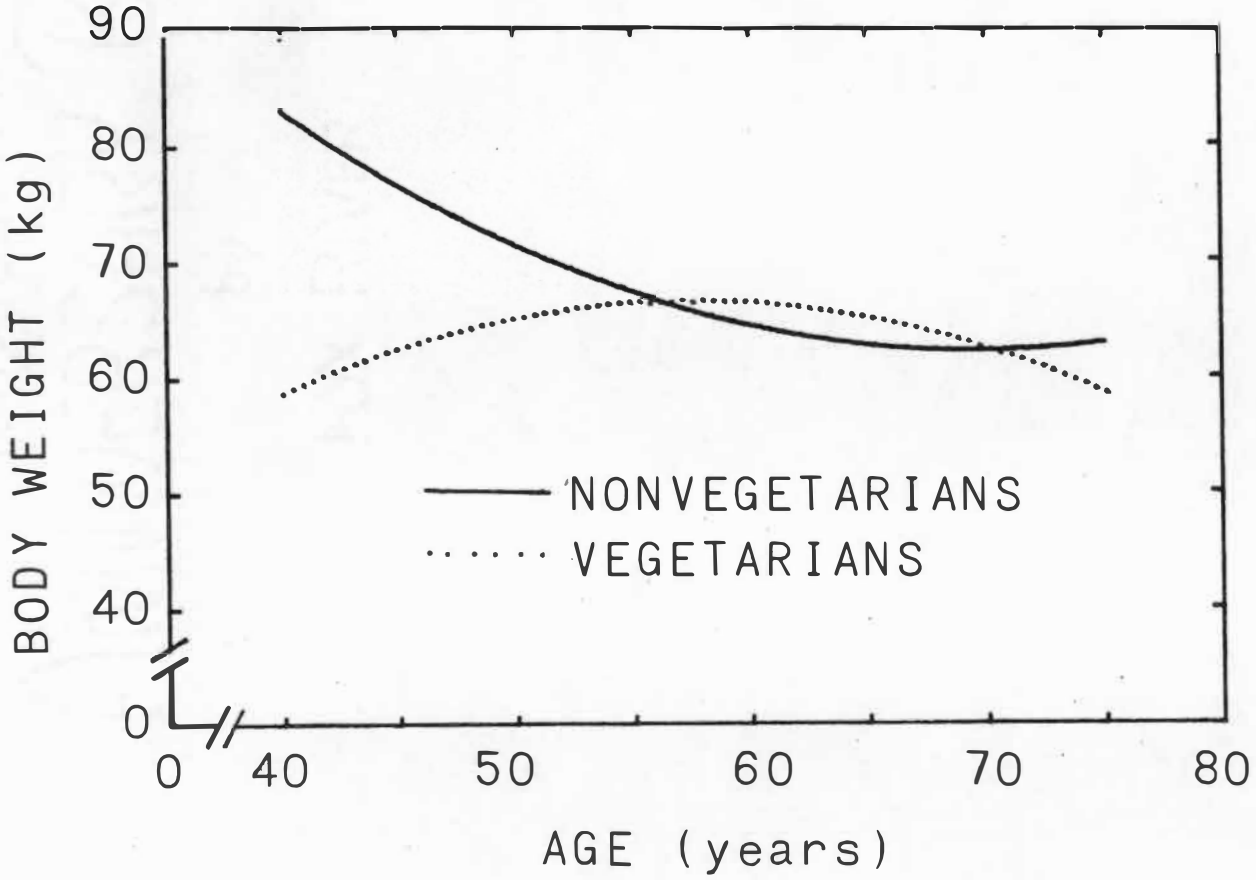


Figure 2. Regression of body weight on age in adult female vegetarians and nonvegetarians. Body weight: nonvegetarians = 177.07 - 3.30 (age) + 0.02 (age$^2$); vegetarians = - 18.78 + 2.97 (age) - 0.02 (age$^2$).

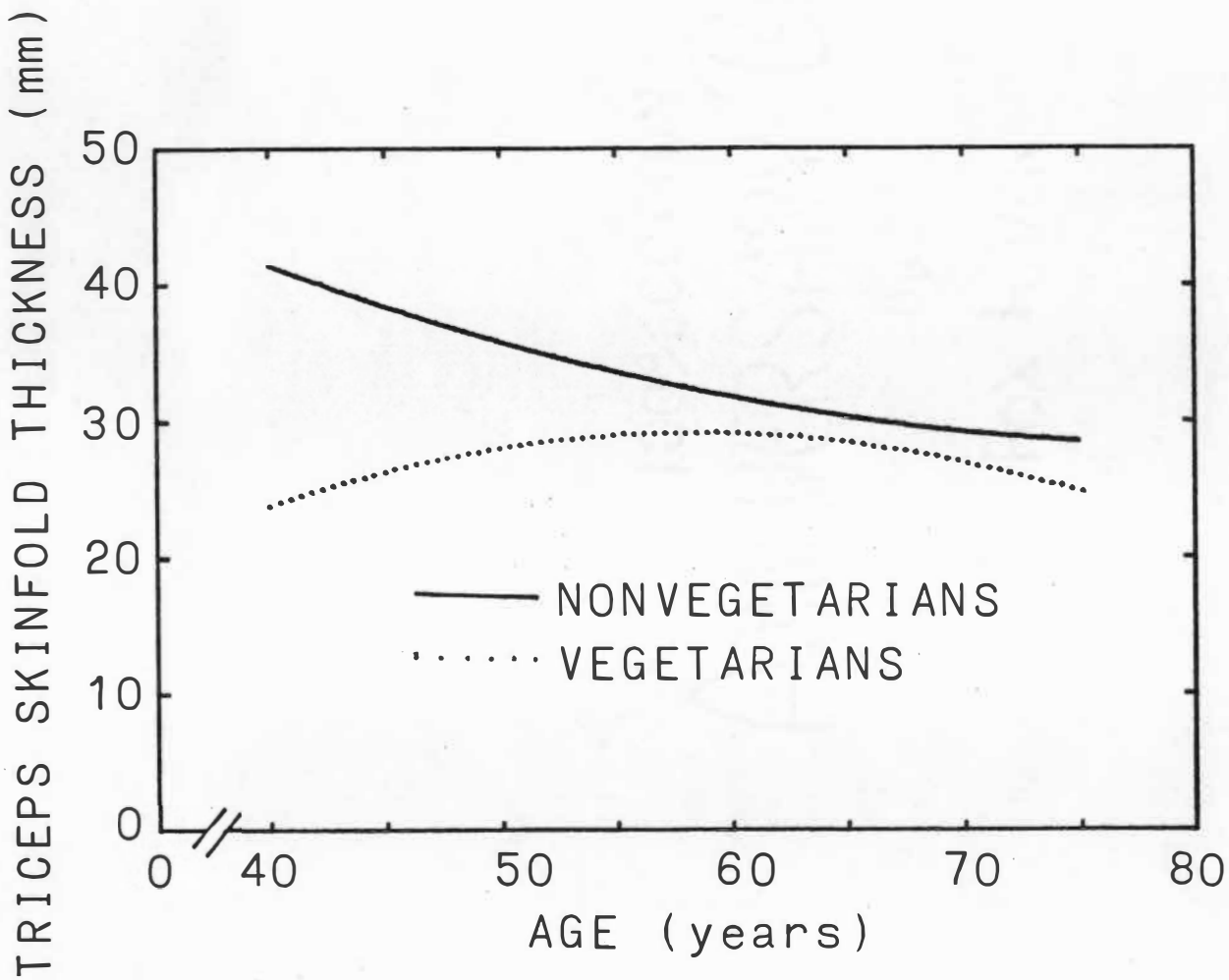


Figure 3. Regression of triceps skinfold thickness on age in adult female vegetarians and nonvegetarians. Triceps skinfold thickness: nonvegetarians = 78.65 - 1.23 (age) + 0.01 ($age^2$); vegetarians = - 24.86 + 1.85 (age) - 0.02 ($age^2$).

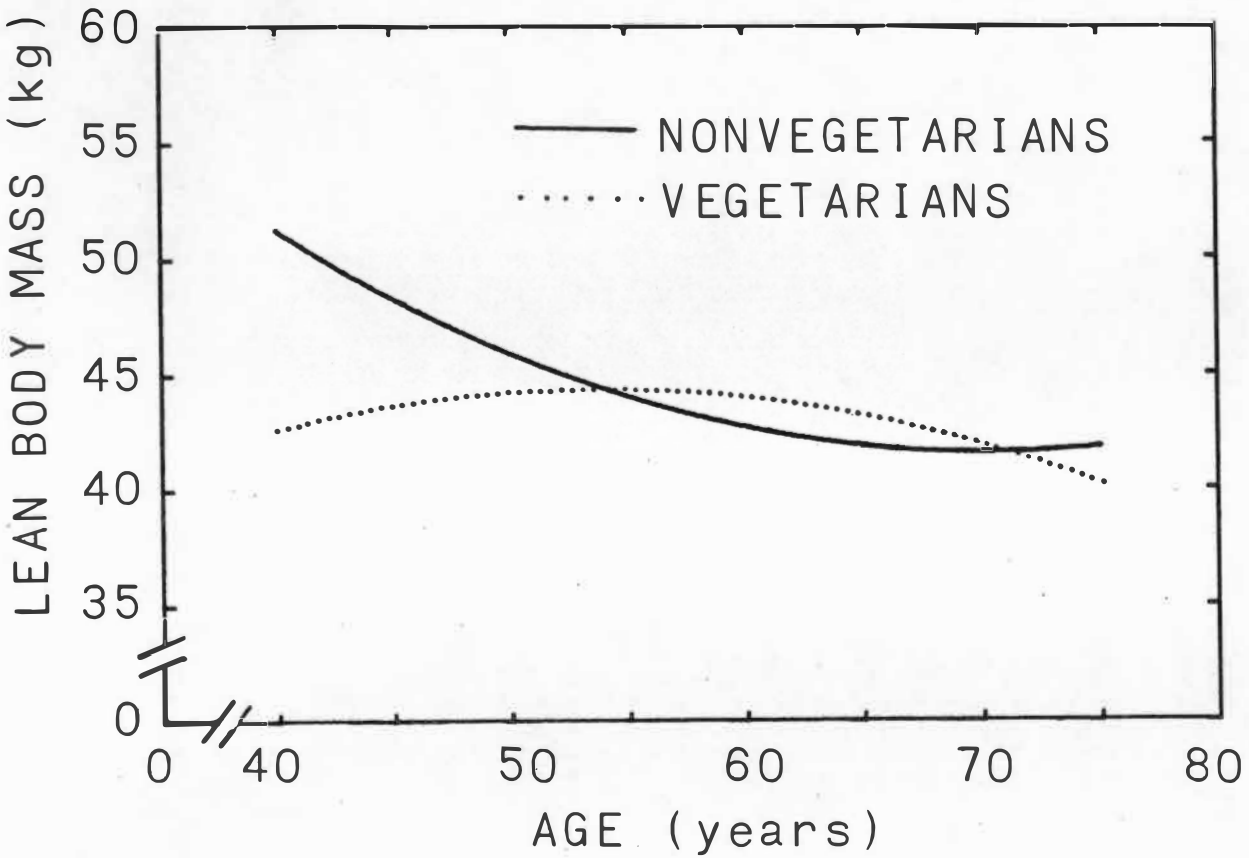


Figure 4. Regression of lean body mass on age in adult female vegetarians and nonvegetarians. Lean body mass: nonvegetarians = 93.87 - 1.49 (age) + 0.01 ($age^2$); vegetarians = 18.19 + 0.97 (age) - 0.01 ($age^2$).

TABLE VII

ANALYSIS OF VARIANCE OF DIETARY INTAKE OF ENERGY AND PROTEIN IN OLDER ADULT FEMALE VEGETARIANS AND NONVEGETARIANS

| Variable | df | Mean Squares | | | |
|---|---|---|---|---|---|
| | | Energy (kcal/day) | Total Protein (g/day) | Animal Protein (g/day) | Vegetable Protein (g/day) |
| Type[1] | 1 | 3996.9 | 4478.4*** | 28283.1*** | 10291.2*** |
| Age | 1 | 1076153.7** | 684.9* | | |
| $Age^2$ | 1 | 77233.5 | 2.1 | | |
| $Age^3$ | 1 | 51866.4 | 22.4 | | |
| Age*Type | 1 | 157221.9 | 0.4 | | |
| $Age^2$*Type | 1 | 72224.7 | 0.9 | | |
| $Age^3$*Type | 1 | 599457.8 | 835.8* | | |
| Residual | | 104568.5 | 149.7 | 163.4 | 67.7 |
| $R^2$ | | 0.14 | 0.26 | 0.59 | 0.56 |

[1]Denotes vegetarian and nonvegetarian diet effects.

*P < 0.05.

**P < 0.01.

***P < 0.001.

Figure 5 is a plot of the regression lines for this relationship. The AV showed a gradual decrease in caloric intake throughout the age range studied while that of NV increased during the fifth decade and decreased until age 70 when a tendency for an increase was observed. Protein intake was significantly different between the two groups and was related to the linear function of age and to the interaction term for the cubic function of age with type of diet. Figure 6 depicts the regression lines for the groups. Protein intake in the AV gradually decreased with age while the NV increased through the fifth decade, plateaued during the sixth and then decreased. Animal and vegetable protein intake showed a significant difference between AV and NV groups but no age effect at any of the levels studied.

Table VIII presents the ANOVA used in developing the equations for urinary excretion of the metabolites studied on age. Total urinary nitrogen showed a significant relationship with the interaction term for the cubic function of age and the type of diet. Figure 7 shows that there is little change in urinary total nitrogen with age in the AV while in NV it increased during the fifth decade and then decreased. Ammonia nitrogen excretion differed significantly between AV and NV groups and also showed a significant age and type of diet interaction term. Figure 8 plots the regression lines for this relationship. The lines are essentially reverse of each other with the NV ammonia nitrogen excretion increasing until the seventh decade and then slowly decreasing. The AV group showed a decrease through the sixth decade followed by a slight increase. Creatinine excretion also differed

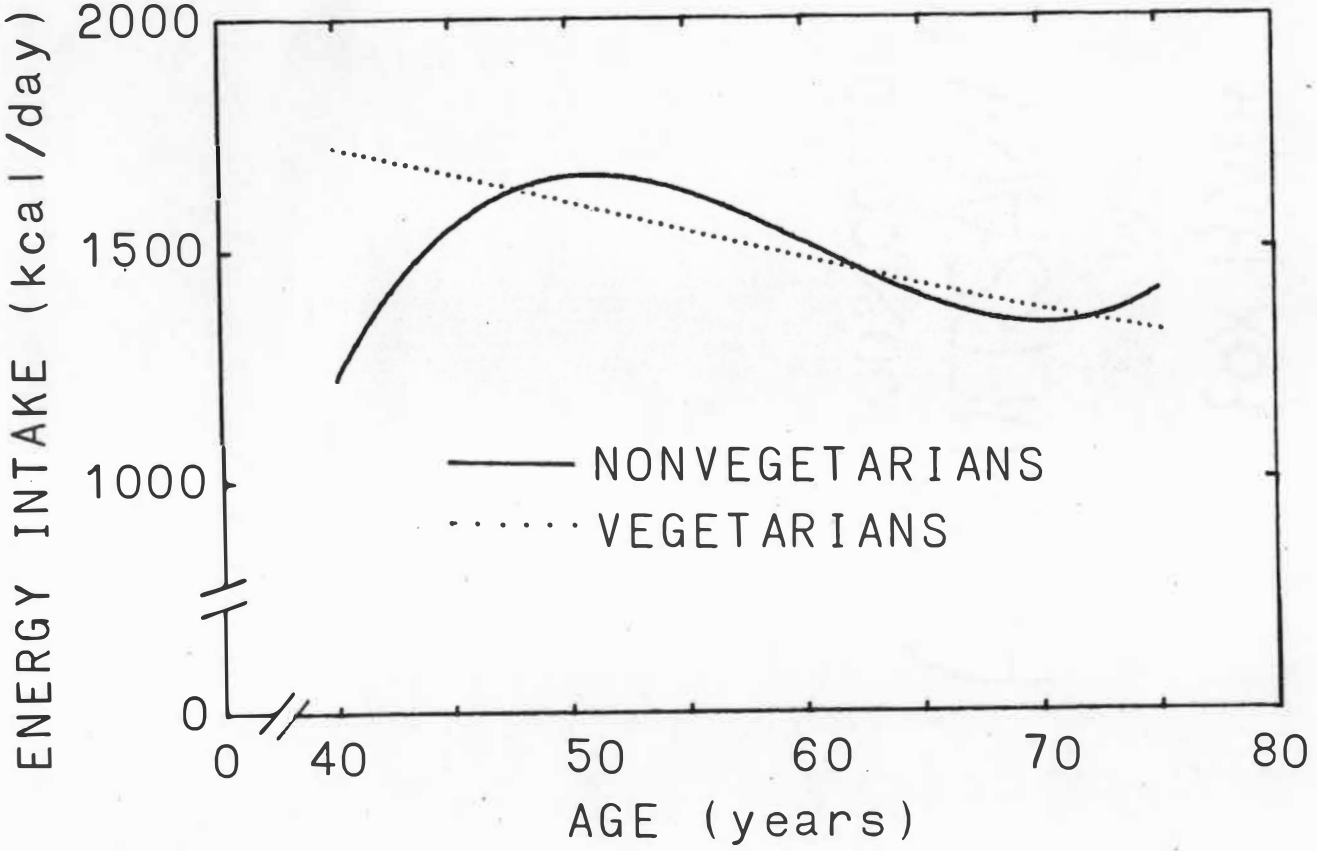


Figure 5. Regression of energy intake on age in adult female vegetarians and nonvegetarians. Energy intake: nonvegetarians = - 17241.91 + 978.05 (age) - 16.55 ($age^2$) + 0.09 ($age^3$); vegetarians = 1818.30 + 10.38 (age) - 0.43 ($age^2$).

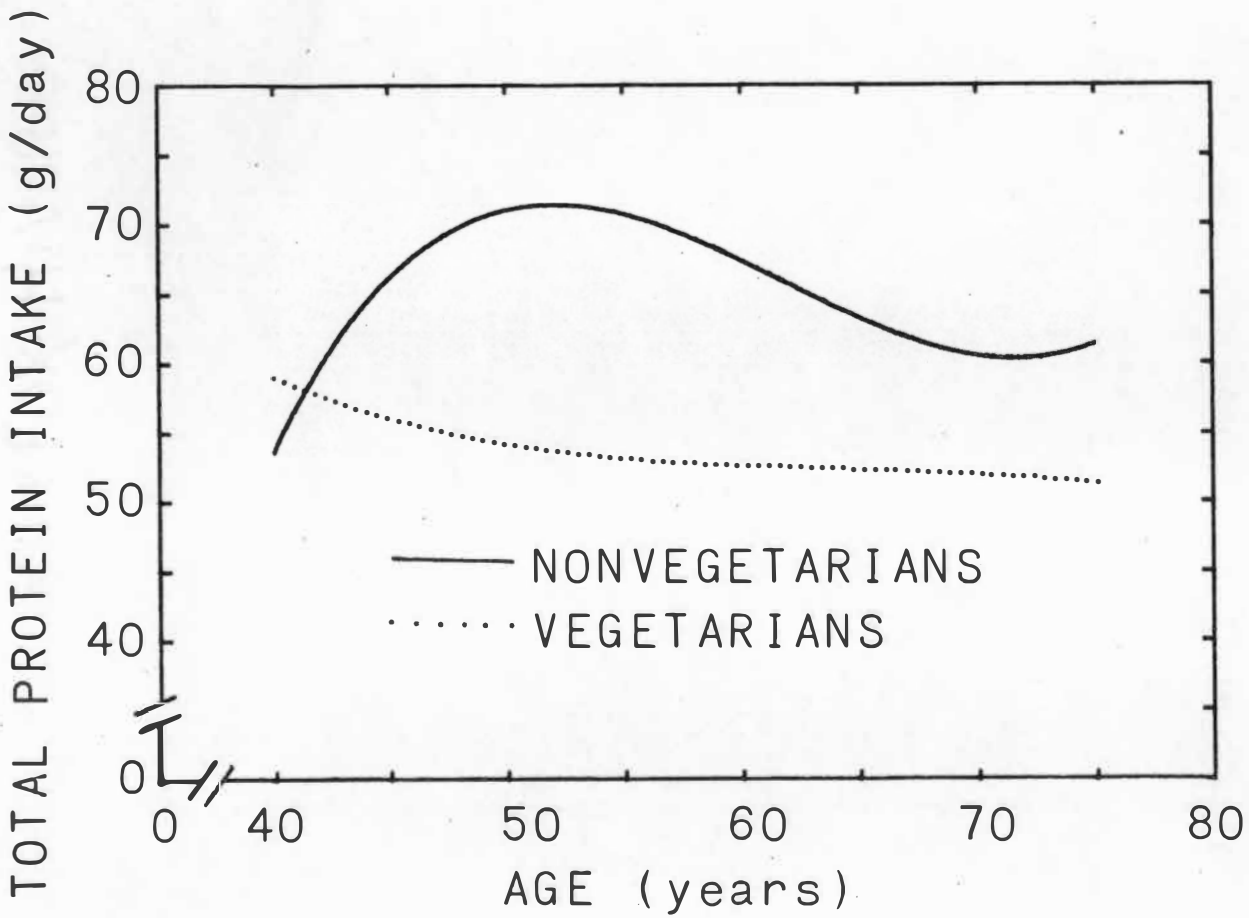


Figure 6. Regression of protein intake on age in adult female vegetarians and nonvegetarians. Protein intake: nonvegetarians = - 576.76 + 32.85 (age) - 0.54 ($age^2$) + 0.01 ($age^3$); vegetarians = 153.44 - 4.60 (age) + 0.07 ($age^2$) - 0.01 ($age^3$).

TABLE VIII

ANALYSIS OF VARIANCE OF EXCRETION OF URINARY COMPONENTS IN OLDER ADULT FEMALE VEGETARIANS AND NONVEGETARIANS

| Variable | df | Mean Squares | | |
|---|---|---|---|---|
| | | Total Nitrogen (g/day) | Ammonia Nitrogen (mg/day) | Creatinine (g/day) |
| Type[1] | 1 | 14182.8 | 95507.2** | 1.41** |
| Age | 1 | 7277.6 | 598.3 | 0.23 |
| $Age^2$ | 1 | 4519.7 | 92.2 | 0.25 |
| $Age^3$ | 1 | 15004.0 | 389.3 | 0.01 |
| Age*Type | 1 | 2418.5 | 70723.0* | 0.77* |
| $Age^2$*Type | 1 | 1503.0 | | |
| $Age^3$*Type | 1 | 29000.2* | | |
| Residual | | 7595.6 | 13709.5 | 0.16 |
| $R^2$ | | 0.08 | 0.09 | 0.12 |

[1]Denotes vegetarian and nonvegetarian diet effects.

*P < 0.05.

**P < 0.01.

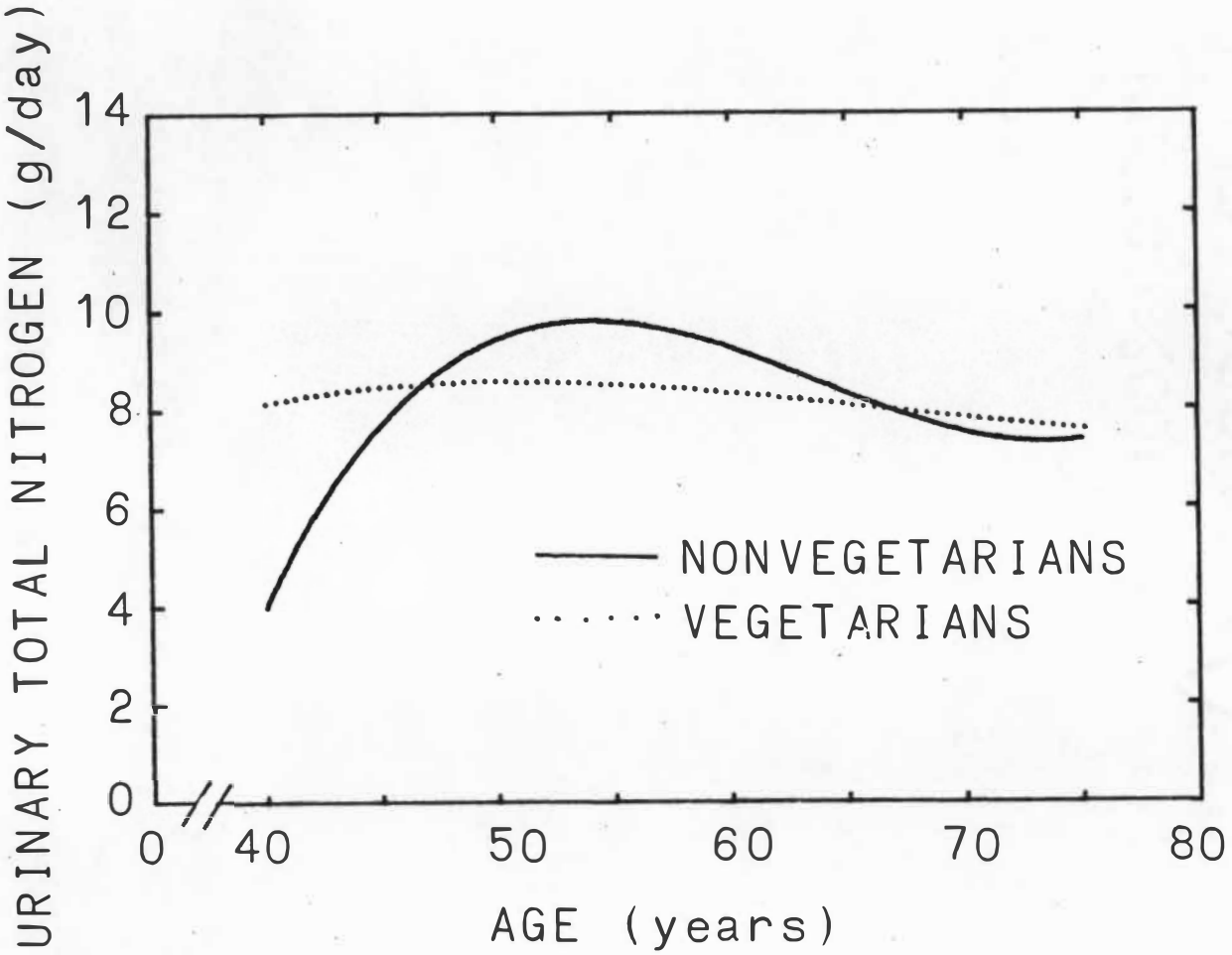


Figure 7. Regression of urinary total nitrogen on age in adult female vegetarians and nonvegetarians. Urinary total nitrogen: nonvegetarians = - 155.44 + 8.10 (age) - 0.13 ($age^2$) + 0.68 ($age^3$); vegetarians = - 8.48 + 0.84 (age) - 0.01 ($age^2$) + 0.07 ($age^3$).

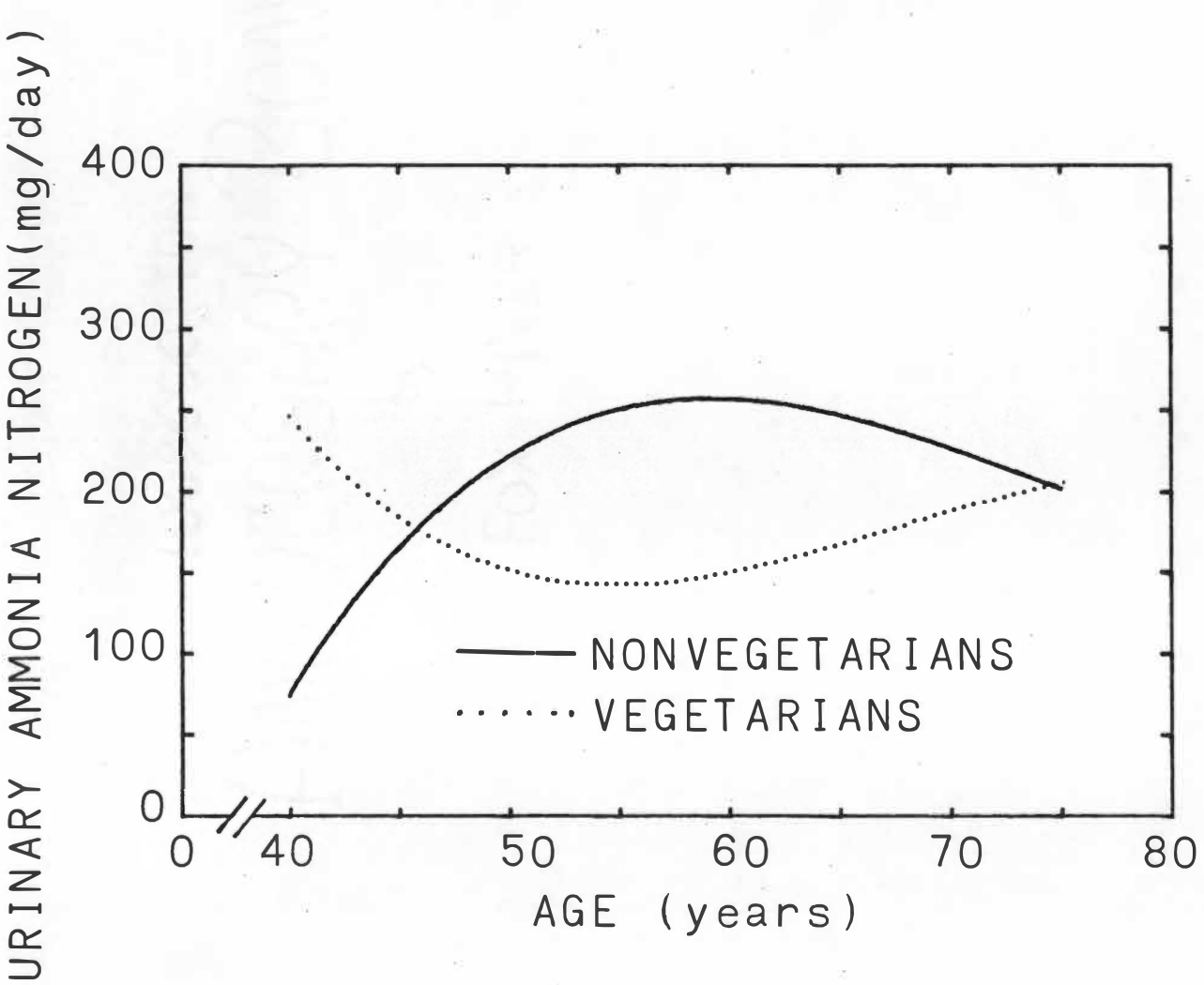


Figure 8. Regression of urinary ammonia nitrogen on age in adult female vegetarians and nonvegetarians. Urinary ammonia nitrogen: nonvegetarians = - 2582.77 = 123.48 (age) - 1.74 ($age^2$) + 0.01 ($age^3$); vegetarians = 2617.32 - 116.87 (age) - 1.79 ($age^2$) - 0.01 ($age^3$).

significantly between the groups, and showed a significant age and type of diet interaction term. Creatinine excretion in the NV exhibited a tendency to increase with age while that of the AV was lower and showed a tendency to decrease with age (Figure 9).

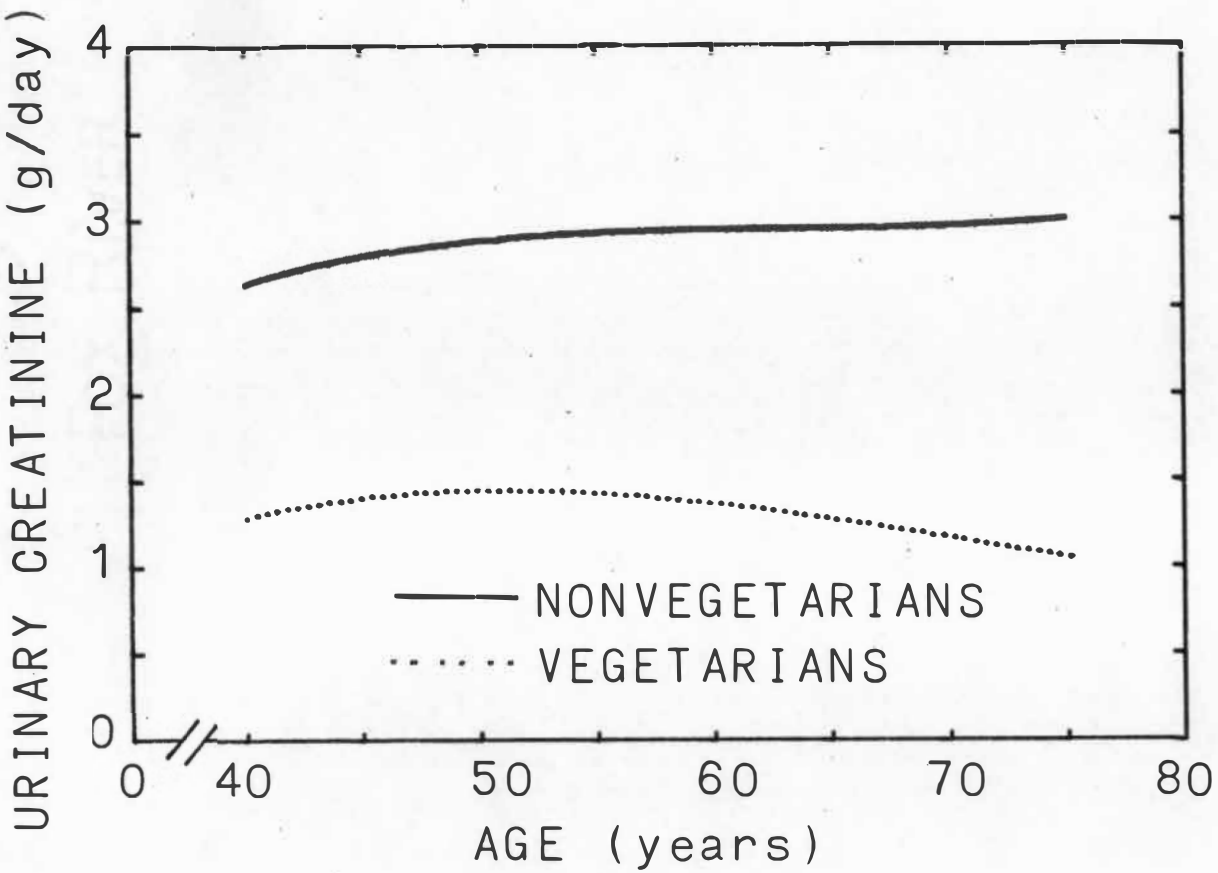


Figure 9. Regression of urinary creatinine on age in adult female vegetarians and nonvegetarians. Urinary creatinine: nonvegetarians = - 3.25 + 2.94 (age) - 0.01 ($age^2$); vegetarians = - 3.92 + 2.58 (age) - 0.02 ($age^2$).

# CHAPTER V

# DISCUSSION

## I. PHYSICAL MEASUREMENTS

Mean height of the AV and NV groups agreed closely with that reported by Durnin and Womersley (41) for British females aged 40 to 68 years and with the results of the Ten-State Nutrition Survey (23). Mean body weight of the V was less while those for LOV and NV were similar to that reported in other studies on females of comparable age (41, 39). Mean triceps skinfold thickness for the LOV and NV were greater than both the 24 mm mean value reported by Durnin and Womersley (41) and the 25.1 mm value used as a standard for the Ten-State Nutrition Survey (23). Using the Ten-State Nutrition Survey (23) standards, 57% of the AV and 77% of the NV were obese. This is much greater than the 30 to 40% found in the Ten-State Nutrition Survey (23). Using the higher 29 mm triceps skinfold thickness standard as suggested by Seltzer and Mayer (44), 38% of the AV and 64% of the NV were still classified as obese. Among the V, 1 of the 6 subjects had a triceps skinfold thickness greater than 25.1 mm indicating a lower incidence of obesity in this group than in LOV and NV and than subjects from other studies (23, 44, 41). Forbes and Reina (37) reported a mean weight of 68 kg, a mean LBM of 38 kg and a mean percent body fat of 45% for females over 40 years of age. The LBM observed in this study for all subjects was slightly greater while

body weight and percent body fat values were less than those found by Forbes and Reina (37).

## II. ENERGY INTAKE

The mean energy intakes for all groups of subjects were less than the RDA (12) with 14% of the AV and 13% of the NV consuming less than 67% of the RDA (1200 kcal/day). The mean energy intakes were also below the 1680 kcal standard used for the Ten-State Nutrition Survey (23) and the 1650 kcal standard used for the USDA Household Food Consumption Survey (14). Most researchers have reported mean energy intakes of the elderly that were below the RDA (12); the most notable exception to these findings was the data from HANES (15). Energy intake data reported in the present study were similar to those obtained by Thompson[1] in these laboratories on elderly nonvegetarian women. Also in agreement with the data of Thompson,[2] the mean weights for the AV and NV groups were 5 to 9 kg more than that of the RDA "reference woman" (12). The present data indicated an inconsistency between the recommendations for caloric intake and body weight in this segment of the population. The RDA (12) is based on a light activity pattern which may overestimate caloric expenditure for the elderly who tend to be rather sedentary. Mean body weight and mean energy intake for the AV subjects were about 10 kg and 150 kcal higher, respectively, than those reported by Brown and Bergan (31) for 23 to 50 year old female vegetarians. All groups of subjects

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[1] Thompson, op. cit.

[2] Ibid.

in the present study had similar body weights and 900 kcal lower energy intakes than those found by Hardinge et al. (33) for vegetarian and nonvegetarian adult females.

## III. PROTEIN INTAKE

All groups of subjects in this study had mean protein intakes which were greater than the allowances recommended by the National Research Council (12) or the Food and Agriculture Organization of the United Nations (13), regardless of whether the intakes were expressed on a g per day or on a g per kg of body weight per day basis. The LOV had a mean daily protein intake which was less than the 1 g per kg of body weight standard used for the HANES (15) and the Ten-State Nutrition Survey (23) and also less than the 0.9 g per kg of body weight standard used for the USDA Household Food Consumption Survey (14). Mean daily protein intakes for the V and NV were greater than these standards. Protein intakes for all groups were less than those reported by Hardinge et al. (33) for nonvegetarian, lactoovovegetarian and vegan females of an age group comparable to that of the subjects in this study. Protein intakes of the vegetarian subjects were greater than those reported by Brown and Bergan (31) for female vegetarians aged 23 to 50 years. Only 19% of the AV and 2% of the NV consumed less than the 46 g per day RDA (12). These percentages are less than those found in the Ten-State Nutrition Survey (23) and in some of the smaller studies (16, 18, 20, 21). The AV derived 15% and the NV, 18% of their total calories from protein which are greater than either the 12% recommended by Harper (25)

or the 12 to 14% recommended by Munro and Young (85). The intake of calories from protein by AV agrees closely with the recommendation of the Senate Select Committee on Nutrition and Health Needs in their Dietary Goals for the United States (86) while that of the NV exceeds these recommendations.

## IV. INTERPRETATION OF THE URINARY DATA

Even though the AV group consumed significantly less protein than the NV, there was no significant difference in the excretion of total urinary nitrogen between the groups. Nitrogen has been reported to be more efficiently utilized on a vegetarian than on a nonvegetarian diet (34) and to be relatively unaffected by age (8). Assuming that multiplying urinary nitrogen by 6.25 will give protein equivalency of that excretion, the AV excreted 97% and the NV, 87% of their dietary protein in the urine. Using obligatory urinary nitrogen losses, the "safe" level of protein intake for elderly women has been reported by Scrimshaw et al. (3) to be 0.42 g per kg of body weight per day. Individuals in nitrogen balance consuming this level of protein would theoretically excrete 67 mg of urinary total nitrogen per kg of body weight per day. The NV consumed 243% of this "safe" level and excreted 200% of the estimated urinary total nitrogen. The AV consumed 210% of the "safe" level and excreted 200% of the estimated total nitrogen. A 13% discrepancy between total protein intake and urinary total nitrogen observed in the NV may be due to a greater retention of nitrogen. The apparent differences between AV and NV in terms of nitrogen retention

were not observed when the data were adjusted to the mean protein intake of the sample. These data can be compared to those of Oddoye and Margen (87) who fed young males a high protein diet for 50 days. The subjects maintained distinctly positive nitrogen balances with no significant trend toward adaptation to this diet which was much higher in protein than the NV diet of the present study. No significant correlation was found between protein intake and nitrogen excretion in the NV apparently due to retention of ingested nitrogen. The difficulty in calculating precise protein intakes for the AV subjects using the recorded food intakes and the available food consumption data may partially explain the low, yet significant, correlation coefficient between protein intake and nitrogen excretion. Expressing the daily protein intake as a mean for 7 days and the urinary data as a single 24-hour sample collected during the week of keeping the dietary record may have resulted in a decrease in the correlation coefficients. In studies in which highly significant correlation coefficients have been obtained between urinary total nitrogen excretion and protein intake, actual protein intake as well as urinary nitrogen excretion have been determined by chemical analyses (3, 4, 24, 34, 8).

The mean creatinine excretions by the AV and NV groups were less than the 1.90 g per day reported by Crim et al. (57, 58) on adult males and by Bleiler and Schedl (59) on adult males and females. The urinary creatinine data were similar to the 1.08 and 1.35 g per day reported for elderly males on protein-free diets (5) and to the 1.14 g per day for adult females on self-chosen diets (40). Other researchers have reported

values which were less than the mean values observed in this study (24). The lower creatinine in the LOV and V as compared to the NV may reflect dietary influences upon the excretion of this compound. Crim et al. (57, 58) reported that the body creatine pool and, hence, creatinine excretion are lowered by feeding a creatine-free (nonmeat) diet. Intakes of both total and animal protein as well as LBM were significantly correlated with creatinine excretion in the LOV; however, in the NV animal protein intake was the only 1 of these 3 variables which was correlated with creatinine excretion. This would seem to indicate that creatinine excretion in the NV was affected by the animal protein intake sufficiently to mask the correlation with LBM. The lack of a significant correlation coefficient between creatinine excretion and LBM or total protein intake in the V group may be due to the small sample size. In the V, creatinine excretion should reflect endogenous creatine metabolism. Adjusting the data for differences in protein intake shows that creatinine excretion in the vegetarian groups is significantly lower than in the NV again indicating the influence of the type of protein ingested. Lin et al. (34) noted creatinine excretions in female vegetarians which were lower than that in nonvegetarians which they attributed to a low protein reserve and protein turnover as a result of adaptation to a prolonged low protein intake.

The lower ammonia nitrogen excretion by LOV compared to NV may reflect differences in protein intake per kg of body weight. A greater intake of protein per kg of body weight would necessitate the increased use of ammonium ion as a urinary buffer for the acid produced in the

catabolism of that protein (88). The ammonia nitrogen excretion by V group did not differ significantly from that of either the NV or LOV groups. This may be because protein intake per kg of body weight for the V was similar to NV and higher than LOV while total protein intake of V was similar to that of LOV and less than that of NV.

The mean HOP excretion by the groups AV and NV were in agreement with that reported by Allison et al. (68), Saleh and Coenegracht (67) and Thompson[3] for older adults. The significantly lower HOP excretion by the V compared to NV may reflect dietary influences or the differences in body weight or in sample size. Dietary HOP is largely of animal origin and is thought to be excreted quantitatively in the urine (64). Thus, HOP excretion by the V would represent endogenous collagen metabolism. The slightly larger HOP excretion in the LOV compared to V, as well as the significant correlation between HOP excretion and animal protein intake in LOV, may indicate consumption of at least some HOP possibly in such dairy products as ice cream and cheese or may reflect the difference in sample size. The V excreted significantly less HOP than the NV even when the data were adjusted to the mean protein intake of the sample. This further illustrated the effect of type of protein consumed.

Urinary inorganic sulfate values for the AV and NV compared favorably with those reported by most other researchers for adult subjects (55, 56, 89). Bodwell et al. (56) found that inorganic sulfate

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[3]Thompson, op. cit.

excretion was increased by feeding adult males diets which either were limiting in one or more of the indispensable amino acids or contained high levels of sulfur amino acids. However, Sabry et al. (90) noted that urinary sulfate excretion was greatly lowered by feeding a rice and beans diet compared to a bread and eggs diet. The urinary sulfate values obtained in the Sabry et al. (90) study were much less than those observed in the present study. The AV seemed to be excreting normal or average amounts of inorganic sulfate, even though they were consuming diets which have the potential for being limiting in the sulfur amino acids. There was no difference in sulfate excretion between the V and LOV or NV even though the V diet could be assumed to resemble the rice and beans diet of Sabry et al. (90). The textured vegetable protein products which were consumed extensively by the vegetarian subjects in this study are frequently fortified with the limiting amino acid or contain egg albumen as a binding agent.[4, 5]

Significant correlations between protein quality and various ratios of urinary metabolites have been reported by many researchers (50, 51, 52, 53, 54, 56, 61). As a result, it has been postulated that determining these ratios may be an accurate and easily obtainable estimate of quality of the protein consumed by a population. Based on extent and significance of correlation with protein intake, the S/N ratio appeared to be the most accurate predictor of protein nutritive value in the LOV. The lack of a significant correlation between any single

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[4]Thompson, op. cit. [5]Ibid.

ratio and protein intake in all the groups agreed with the conclusions of Swendseid and Wang (51), and Bodwell et al. (56), i.e., in the presence of adequate protein intake, multiple parameter indices of protein nutritive value are necessary.

## V. AGE RELATIONSHIPS

The cross-sectional data reported in this study cannot be interpreted to represent age changes for any given individual. They do, however, indicate age differences among women of varying ages at a given time. It has been generally accepted that age-associated decrements in physiological functions occur as a linear function of age (91). In the present study, polynomial regression models were frequently found to more accurately describe the relationship between age and a variable. In all instances, the regression lines were not continued beyond age 75 years because the small number of subjects past this age contributed to the inherent tendency of a polynomial to magnify variations at either extreme of the data.

Height decreased linearly with age in both the AV and the NV. Body weight in the NV tended to decrease with increasing age. In contrast, the results for the AV showed that women aged 50 to 60 years had a greater body weight than women aged 40 to 50 years or over 60. The latter data on the AV subjects agreed closely with those found in the Ten-State Nutrition Survey (23). LBM was found to follow the same basic pattern as body weight in both groups. However, creatinine excretion showed essentially no change in the AV with increasing age and slightly

increased in the NV. This was perhaps due to the increased breakdown of muscle protein relative to total body protein with increasing age, as found by Young and coworkers (1, 45). They have reported that body composition changes with age and that the visceral tissue comprises a greater percentage of total body protein in the elderly than in younger adults. Therefore, one would expect greater creatinine excretion with increasing age due to muscle protein catabolism. Skinfold thickness and percent body fat decreased with increasing age in the NV and increased through age 60 in the AV. As with body weight, the skinfold thickness and body fat data of AV followed the same pattern as that of the Ten-State Nutrition Survey (23). The fact that body weight, LBM and skinfold thickness (or percent body fat) exhibited the same trends with increasing age within a group would seem to indicate that changes in each of the body components with age were parallel.

Energy and protein intake in the AV showed a gradual decrease with age. The NV showed an increase in energy and protein intake during the sixth decade followed by a decrease in the seventh decade and a tendency to increase in later life. These variations may be due to sampling bias. The 50 to 59 year old subjects represent an active group who consumed rather large quantities of food. There were also a few subjects in the oldest grouping who were very active and consumed large quantities of food. The women in this oldest age group may represent a statistically biased sample because of the small number of subjects as well as the few people who live to this age. Thus, it is not possible to say whether these women were unusual or whether they actually represented the few

people who live to this age. The validity of using dietary records to assess nutrient intake in the elderly has been questioned (24). However, the dietary intake data of this study were supported by the total nitrogen excretion data. That is, the age segments of the various groups who had large protein intakes also had greater urinary nitrogen excretions.

The lack of a significant relationship between age and urinary inorganic sulfate or urea nitrogen may indicate that dietary protein metabolism did not change within the age range studied. It may also support the apparent adequacy of the protein intake for all the age groups. HOP excretion remained essentially constant across the age span of this study which agreed with the data of Allison et al. (68) and Williams and Windsor (69). The lack of change in HOP excretion may be due to the increased catabolism of collagen which would accompany declines in bone mass and in LBM with age.

The most notable differences observed between the AV and NV subjects were in body fat (skinfold thickness), amount and source of dietary protein and in the urinary excretion of creatinine. The AV and NV were similar in height and urinary excretion of HOP and inorganic sulfate throughout the age range studied. The 2 groups were also very similar between the ages of 50 and 70 years with regard to body weight, LBM and caloric intake. In this age range, the groups were somewhat less similar in their body fat (skinfold thickness) and urinary ammonia and nitrogen excretion. The nitrogen excretion for each group followed the same trend with age as the protein and energy intakes. However, the protein

intake was significantly different between the 2 groups while nitrogen excretion was not. Creatinine excretion was also significantly different for the 2 groups. The V subjects had a lower body weight, body fat (skinfold thickness), LBM and urinary HOP than the NV and tended to be lower than LOV in these parameters. However, due to the small number of subjects in the V group, no definitive conclusions may be drawn. Thus, the consumption of a vegan diet may have an effect on selected physical measurements and the intake and metabolism of protein; the consumption of a lactoovovegetarian diet has a much smaller effect.

# CHAPTER VI

## SUMMARY

The mean heights of the V, LOV and NV did not differ significantly from each other nor did the AV and NV differ in the regression of height on age. The V (51.0 ± 5.6 kg) weighed significantly less than the LOV (65.8 ± 1.8 kg) and NV (66.7 ± 1.6 kg). LBM for the V (39.9 ± 2.1 kg) was also significantly less than that of the LOV (43.9 ± 0.7 kg) and NV (43.7 ± 0.6 kg). Percent body fat (skinfold thickness) for V (14.8 ± 3.9%) was significantly less than that of LOV (24.1 ± 1.2%); both vegetarian groups were significantly less fat than NV (28.1 ± 1.1%). There was a significant decrease in weight, LBM and percent body fat with age in both AV and NV as well as significant differences in the regression of these physical measurements on age. The groups were remarkably similar with respect to these values between ages 50 and 70 years. Within each group there were parallel changes in body weight, LBM and percent body fat throughout the age range of the data.

Mean energy intakes for the V, LOV and NV groups did not differ. However, there was a significant decrease in caloric consumption with age. Protein intake also decreased with age and differed significantly between the LOV and V and the NV. The values were 54.6 ± 1.3, 52.2 ± 4.2 and 66.5 ± 1.6 g/day, respectively. Consumption of animal and vegetable protein differed significantly between the vegetarian and nonvegetarian groups; the differences were not age-associated. The NV consumed 45.8 ± 1.7 g of animal protein, significantly more than the 16.0 ± 1.7 g

consumed by the LOV. The V consumed 38.2 ± 3.2 g of vegetable protein/day which was significantly more than the 28.2 ± 1.1 g consumed by the LOV. Both vegetarian groups consumed significantly more vegetable protein than the NV (10.8 ± 1.1 g/day).

Although mean urinary total nitrogen excretion did not differ among the groups, it did exhibit the same trends with age as that of protein and energy intake. There were no significant differences in urinary urea nitrogen or inorganic sulfate excretion among the groups. HOP excretion by the V (28.3 ± 4.8 mg/day) was significantly less than that of the NV (35.5 ± 1.7 mg); the 33.2 ± 1.6 mg/day excreted by the LOV did not differ significantly from that of either other group. Urinary excretion of urea nitrogen, inorganic sulfate and HOP were not related to age at any of the levels studied. The V (1.30 ± 0.14 g) and LOV (1.33 ± 0.04 g) excreted significantly less creatinine than the NV (1.54 ± 0.05 g/day). Changes in creatinine excretion with age differed between the AV and NV groups. Adjusting the urinary data to the mean protein intake of the sample did not change the patterns of significance.

Based on the urinary nitrogen excretion in relation to protein intake, it appears that NV were retaining more of the ingested protein than the AV. Since changes in total nitrogen excretion with age were parallel to changes in protein intake with age, the observed differences in retention for AV and NV would seem to be consistent throughout the age span studied. The AV and NV groups differed primarily in body fat, in the amount and nature of the dietary protein and in the urinary excretion of creatinine. The V subjects had a lower body weight, body

fat, LBM and urinary HOP than the NV and tended to be lower than the LOV. However, the small number of V subjects precludes drawing definite conclusions. The observed differences and similarities among the V, LOV and NV indicate that the consumption of a vegan diet may have a more extensive effect on physical measurements and intake and metabolism of protein than the consumption of a lactoovovegetarian diet.

# LITERATURE CITED

## LITERATURE CITED


1. Young, V. R., Perera, W. D., Winterer, J. C. & Scrimshaw, N. S. (1976) Protein and amino acid requirements of the elderly. In: Nutrition and Aging (Winick, M., ed.), pp. 77-118, John Wiley & Sons, Inc., New York.

2. Winick, M. (1978) The relationship of diet, nutrition and health during various stages of life. Food Technol. 32, 42-43.

3. Scrimshaw, N. S., Perera, W. D. A. & Young, V. R. (1976) Protein requirements of man: Obligatory urinary and fecal nitrogen losses in elderly women. J. Nutr. 106, 665-670.

4. Bricker, M. L. & Smith, J. M. (1951) A study of endogenous nitrogen output of college women, with particular reference to use of the creatinine output in the calculation of the biological values of the protein of egg and sunflower seed flour. J. Nutr. 44, 553-573.

5. Uauy, R., Scrimshaw, N. S., Rand, W. R. & Young, V. R. (1978) Human protein requirements: Obligatory urinary and fecal nitrogen losses and the factorial estimation of protein needs in elderly males. J. Nutr. 108, 97-103.

6. Zanni, E., Calloway, D. H. & Zezulka, A. Y. (1979) Protein requirements of elderly men. J. Nutr. 109, 513-524.

7. Calloway, D. H. & Margen, S. (1971) Variation in endogenous nitrogen excretion and dietary nitrogen utilization as determinants of human protein requirement. J. Nutr. 101, 205-216.

8. Cheng, A. H. R., Gomez, A., Bergan, J. G., Lee, T-C., Monckeberg, F. & Chichester, C. O. (1978) Comparative nitrogen balance study between young and aged adults using 3 levels of protein intake from a combination wheat-soy-milk mixture. Am. J. Clin. Nutr. 31, 12-22.

9. Tuttle, S. G., Bassett, S. H., Griffith, W. H., Mulcare, D. B. & Swendseid, M. E. (1965) Further observations on the amino acid requirements of older men. II. Methionine and lysine. Am. J. Clin. Nutr. 16, 229-231.

10. Watts, J. H., Mann, A. N., Bradley, L. & Thompson, D. J. (1964) Nitrogen balances of men over 65 fed the FAO and milk patterns of essential amino acids. J. Gerontol. 19, 370-374.

11. Tontisirin, K., Young, V. R., Miller M. & Scrimshaw, N. S. (1973) Plasma tryptophan response curve and tryptophan requirements of elderly people. J. Nutr. 103, 1220-1228.

12. Food and Nutrition Board, National Research Council (1974) Recommended Dietary Allowances, ed. 8, National Academy of Sciences, Washington, D.C.

13. Report of a Joint FAO/WHO Ad Hoc Expert Committee (1973) Energy and protein requirements. World Health Organization Technical Report Series, No. 522, WHO, Geneva.

14. Consumer and Food Economics Research Division, Agricultural Research Service (1972) Food and nutrient intake of individuals in the United States, Spring, 1965. Household Food Consumption Survey, Report No. 11, U.S. Government Printing Office, Washington, D.C.

15. Preliminary Findings of the First Health and Nutrition Examination Survey, U.S., 1971-1972 (1974) Dietary intake and biochemical findings. DHEW Publication No. (HRA) 74-1219-1. U.S. Government Printing Office, Washington, D.C.

16. Jansen, C. & Harrill, I. (1977) Intakes and serum levels of protein and iron for 70 elderly women. Am. J. Clin. Nutr. 30, 1414-1422.

17. Greger, J. L. & Sciscoe, B. S. (1977) Zinc nutriture of elderly participants in an urban feeding program. J. Am. Dietet. Assoc. 70, 37-41.

18. Jordan, V. E. (1976) Protein status of the elderly as measured by dietary intake, hair tissue, and serum albumin. Am. J. Clin. Nutr. 29, 522-528.

19. Kohrs, M. B., O'Neal, R., Preston, A., Eklund, D. & Abrahams, O. (1978) Nutritional status of elderly residents in Missouri. Am. J. Clin. Nutr. 31, 2186-2197.

20. MacLennan, W. J., Martin, P. & Mason, B. J. (1977) Protein intake and serum albumin levels in the elderly. Gerontol. 23, 360-367.

21. Justice, C. L., Howe, J. M. & Clark, H. E. (1974) Dietary intakes and nutritional status of elderly patients. J. Am. Dietet. Assoc. 65, 639-646.

22. Report by Nutrition Canada to the Dept. of Nat'l. Health and Welfare (1973) Nutrition Canada National Survey, 1970-1972. Information Canada, Ottawa.

23. Center for Disease Control, U.S. Department of Health, Education, and Welfare (1972) Ten-State Nutrition Survey, 1968-1970: I. Dietary. DHEW Publ. No. (HSM) 72-8133, Atlanta, GA.

24. DaCosta, F. & Moorhouse, J. A. (1969) Protein nutrition in aged individuals on self-selected diets. Am. J. Clin. Nutr. 22, 1618-1633.

25. Harper, A. E. (1978) Recommended dietary allowances for the elderly. Geriatrics 33, 78-80.

26. Davidson, C. S., Livermore, J., Anderson, P. & Kaufman, S. (1962) The nutrition of a group of apparently healthy aging persons. Am. J. Clin. Nutr. 10, 181-199.

27. McGandy, R. B., Barrows, C. H., Spanias, A., Meredith, A., Stone, J. L. & Norris, A. H. (1966) Nutrient intakes and energy expenditure in men of different ages. J. Gerontol. 21, 581-587.

28. Harrill, I., Erbes, C. & Schwartz, C. (1976) Observations on food acceptance by elderly women. Gerontol. 16, 349-355.

29. Macleod, C. C., Judge, T. G. & Caird, F. I. (1974) Nutrition of the elderly at home. I. Intakes of energy, protein, carbohydrates and fat. Age & Ageing 3, 158-166.

30. Hardinge, M. G. & Stare, F. J. (1954) Nutritional studies of vegetarians. I. Nutritional, physical and laboratory studies. Am. J. Clin. Nutr. 2, 73-82.

31. Brown, P. T. & Bergan, J. G. (1975) The dietary status of "new" vegetarians. J. Am. Dietet. Assoc. 67, 455-459.

32. Register, U. D. & Sonnenberg, L. M. (1973) The vegetarian diet. J. Am. Dietet. Assoc. 62, 253-261.

33. Hardinge, M. G., Crooks, H. & Stare, F. J. (1966) Nutritional studies of vegetarians. V. Proteins and essential amino acids. J. Am. Dietet. Assoc. 48, 26-28.

34. Lin, T., Chen, M. L. & Chen, J. S. (1973) Observation on dietary protein utilization in vegetarians. Chin. J. Physiol. 21, 143-150.

35. Register, U. D., Inano, M., Thurston, C. E., Vyhmeister, I. B., Dysinger, P. W., Blankenship, J. W. & Horning, M. C. (1967) Nitrogen-balance studies in human subjects on various diets. Am. J. Clin. Nutr. 20, 753-759.

36. Cohn, S. H., Vaswani, A., Zanzi, I., Alioa, J. F., Roginsky, M. S. & Ellis, K. J. (1976) Changes in body chemical composition with age measured by total-body neutron activation. Metabolism 25, 85-95.

37. Forbes, G. B. & Reina, J. C. (1970) Adult lean body mass declines with age: Some longitudinal observations. Metabolism 19, 653-663.

38. Forbes, G. B. (1974) Stature and lean body mass. Am. J. Clin. Nutr. 27, 595-602.

39. Hume, R. (1966) Prediction of lean body mass from height and weight. J. Clin. Pathol. 19, 389-391.

40. Forbes, G. B. & Bruining, G. J. (1976) Urinary creatinine excretion and lean body mass. Am. J. Clin. Nutr. 29, 1359-1366.

41. Durnin, J. V. G. A. & Womersley, J. (1974) Body fat assessed from body density and its estimation from skinfold thickness measurements on 481 men and women aged from 16 to 72 years. Br. J. Nutr. 32, 77-97.

42. Womersley, J. & Durnin, J. V. G. A. (1977) A comparison of the skinfold method with extent of "overweight" and various weight-height relationships in the assessment of obesity. Br. J. Nutr. 38, 271-284.

43. Hill, G. L., Bradley, J. A., Collins, J. P., McCarthy, I., Oxby, C. B. & Burkinshaw, L. (1978) Fat-free body mass from skinfold thickness: A close relationship with total body nitrogen. Br. J. Nutr. 39, 403-405.

44. Seltzer, C. C. & Mayer, J. (1967) Greater reliability of the triceps skin fold over the subscapular skin fold as an index of obesity. Am. J. Clin. Nutr. 9, 950-953.

45. Young, V. R. (1976) Protein metabolism and needs in elderly people. In: Nutrition, Longevity, and Aging (Rockstein, M. & Sussman, M. L., eds.), pp. 67-102, Academic Press, New York.

46. Taylor, Y. S. M., Scrimshaw, N. S. & Young, V. R. (1974) The relationship between serum urea levels and dietary nitrogen utilization in young men. Br. J. Nutr. 32, 407-411.

47. Young, V. R., Fajardo, L., Murray, E., Rand, W. M. & Scrimshaw, N. S. (1975) Protein requirements of man: Comparative nitrogen balance response within the submaintenance-to-maintenance range of intakes of wheat and beef proteins. J. Nutr. 105, 534-542.

48. Prior, R. L., Milner, J. A. & Visek, W. J. (1975) Urea, citrate and orotate excretions in growing rats fed amino acid-deficient diets. J. Nutr. 105, 141-146.

49. Brown, J. A. & Cline, T. R. (1974) Urea excretion in the pig: An indicator of protein quality and amino acid requirements. J. Nutr. 104, 542-545.

50. Nakagawa, I. & Masana, Y. (1967) Assessment of nutritional status of men: Protein. J. Nutr. 93, 135-141.

51. Swendseid, M. E. & Wang, M. (1970) Sulfur in human nutrition. In: Sulfur in Nutrition (Muth, O. H. & Oldfield, J. E., eds.), pp. 209-221, The AVI Publishing Co., Inc., Westport, CT.

52. Bodwell, C. E. (1977) Biochemical indices in humans. In: Evaluation of Proteins for Humans (Bodwell, C. E., ed.), pp. 119-148, The AVI Publishing Co., Inc., Westport, CT.

53. Miller, D. S. & Mumford, P. (1964) Urinary sulphur as a measure of the protein value of diets. Proc. Nutr. Soc. 23, 44-46.

54. Pellett, P. L. (1965) Urinary sulphate sulphur as a measure of the protein value of diets. Proc. Nutr. Soc. 24, 37-39.

55. Lakshmanan, F. L., Perera, W. D. A., Scrimshaw, N. S. & Young, V. R. (1976) Plasma and urinary amino acids and selected sulfur metabolites in young men fed a diet devoid of methionine and cystine. Am. J. Clin. Nutr. 29, 1367-1371.

56. Bodwell, C. E., Schuster, E. M., Brooks, B. & Womack, M. (1978) Biochemical indices in humans of protein nutritive value. I. Urinary inorganic sulfate excretion at a high protein intake level. Nutr. Rep. Intl. 18, 125-133.

57. Crim, M. C., Calloway, D. H. & Margen, S. (1975) Creatine metabolism in men: Urinary creatine and creatinine excretion with creatine feeding. J. Nutr. 105, 428-438.

58. Crim, M. C., Calloway, D. H. & Margen, S. (1976) Creatine metabolism in men: Creatine pool size and turnover in relation to creatine intake. J. Nutr. 106, 371-381.

59. Bleiler, R. E. & Schedl, H. P. (1962) Creatinine excretion: Variability and relationships to diet and body size. J. Lab. Clin. Med. 59, 945-955.

60. Murlin, J. R., Szymanski, T. A. & Nasset, E. C. (1948) Creatinine nitrogen percentage as a check on the biological values of proteins. J. Nutr. 36, 171-175.

61. Powell, R. C., Plough, I. C. & Baker, E. M. (1961) The use of nitrogen to creatinine ratios in random urine specimens to estimate dietary protein. J. Nutr. 73, 47-52.

62. VanNiekerk, B. D. H., Reid, J. T., Bensadoun, A. & Paladines, O. L. (1963) Urinary creatinine as an index of body composition. J. Nutr. 79, 463-473.

63. Duggal, S. K. & Eggum, B. O. (1978) Urinary creatinine and creatine excretion in pigs in relation to body weight and nitrogen balance. J. Sci. Food Agric. 29, 683-688.

64. Meilman, E., Urivetzky, M. M. & Rapoport, C. M. (1963) Urinary hydroxyproline peptides. J. Clin. Invest. 42, 40-49.

65. Jasin, H. E., Fink, C. W., Wise, W. & Ziff, M. (1962) Relationship between urinary hydroxyproline and growth. J. Clin. Invest. 41, 1928-1935.

66. Weiss, P. H. & Klein, L. (1969) The quantitative relationship of urinary peptide hydroxyproline excretion to collagen degradation. J. Clin. Invest. 48, 1-10.

67. Saleh, A. E. C. & Coenegracht, J. M. (1968) The influence of age and weight on the urinary excretion of hydroxyproline and calcium. Clin. Chim. Acta 21, 445-452.

68. Allison, D. J., Walker, A. & Smith, Q. T. (1966) Urinary hydroxyproline:creatinine ratio of normal humans at various ages. Clin. Chim. Acta 14, 729-734.

69. Williams, C. B. & Windsor, A. C. M. (1971) The use of the hydroxyproline:creatinine ratio in elderly patients. Gerontol. Clinica 13, 277-284.

70. Watt, B. K. & Merrill, A. L. (1963) Composition of foods—raw, processed, prepared. Revised. Agric. Handbook No. 8, U.S. Dept. of Agriculture, Washington, D.C.

71. Adams, C. F. (1975) Nutritive Value of American Foods in Common Measures. Agric. Handbook No. 456, U.S. Dept of Agriculture, Washington, D.C.

72. Church, C. F. & Church, H. N. (1975) Food Values of Portions Commonly Used, ed. 12, J. B. Lippincott Company, Philadelphia.

73. Physician's Desk Reference (1974) ed. 28, Medical Economics Company, Oradell, N.J.

74. Tanner, J. M. & Whitehouse, R. H. (1962) Standards for subcutaneous fat in British children. Br. Med. J. 1, 446-450.

75. Dambacher, M., Gubler, A. & Haas, H. G. (1968) A new time-saving method for determining nitrogen in biologic materials. Clin. Chem. 14, 615-622.

76. Mann, L. T. (1963) Spectrophotometric determination of nitrogen in total micro-Kjeldahl digest. Anal. Chem. 35, 2179-2182.

77. Richterich, R. (1969) Clinical Chemistry—Theory and Practice, pp. 253-258, Academic Press, N.Y.

78. Henry, R. J. (1967) Determination of creatinine and creatine. In: Clinical Chemistry: Principles and Techniques (Henry, R. J., ed.), pp. 292-296, Harper and Row, N.Y.

79. Bergman, I. & Loxley, R. (1970) The determination of hydroxyproline in urine hydrolysates. Clin. Chim. Acta 27, 347-349.

80. Hawk, P. B. (1965) Physiological Chemistry, ed. 14 (Oser, B. L., ed.), pp. 1214-1233, McGraw-Hill Book Company, N.Y.

81. Association of Official Analytical Chemists (1970) Official Methods of Analysis, ed. 11 (Horwitz, E., ed.), p. 874, Association of Official Analytical Chemists, Washington, D.C.

82. Swaroop, A. (1973) A micromethod for the determination of urinary inorganic sulfates. Clin. Chim. Acta 46, 333-336.

83. Barr, A. J. (1976) Statistical Analysis System. SAS Institute Inc., Raleigh, N.C.

84. Steele, R. G. D. & Torrie, J. H. (1960) Principles and Procedures of Statistics, pp. 107-109, McGraw-Hill Book Company, Inc., N.Y.

85. Munro, H. N. & Young, V. R. (1978) Protein metabolism in the elderly. Postgrad. Med. 63, 143-149.

86. Select Committee on Nutrition and Human Needs (1977) Dietary Goals for the United States. U.S. Government Printing Office, Washington, D.C.

87. Oddoye, E. A. & Margen, S. (1979) Nitrogen balance studies in humans: Long-term effect of high nitrogen intake on nitrogen accretion. J. Nutr. 109, 363-377.

88. Bhagavan, N. V. (1974) Biochemistry: A Comprehensive Review, pp. 848-858, J. B. Lippincott Company, Philadelphia.

89. Freyberg, R. H., Block, W. D. & Fromer, M. F. (1940) A study of sulfur metabolism and the effect of sulfur administration in chronic arthritis. J. Clin. Invest. 19, 423-435.

90. Sabry, Z. I., Shadarevian, S. B., Cowan, J. W. & Campbell, J. A. (1965) Relationship of dietary intake of sulphur amino-acids to urinary excretion of inorganic sulphate in man. Nature 206, 931-933.

91. Barrows, C. H. & Beauchene, R. E. (1970) Aging and nutrition. In: Newer Methods of Nutritional Biochemistry (Albanese, A. A., ed.), pp. 163-194, Academic Press, N.Y.

# APPENDIXES

# APPENDIX A

UNIVERSITY OF TENNESSEE—KNOXVILLE

TENNESSEE AGRICULTURAL EXPERIMENT STATION

Project Consent Form

I agree, as indicated by my signature below, that:

(1) I would like to participate in the Nutrition and Bone Density Project approved and administered by the professional staff of the Tennessee Agricultural Experiment Station and the College of Home Economics, University of Tennessee, Knoxville;

(2) I understand that this project has been judged by the professional staff as not likely to be harmful to the participants involved or an inappropriate or unnecessary invasion of the privacy of the families;

(3) I understand that participation in this program is not likely to harm me and that no specific benefits or effects as guaranteed other than information from the assessment of my bone density and nutrient intake;

(4) It is my understanding that each aspect of the project in which I am asked to participate will be explained to me and that I may withdraw from participation at any time if involvement is unacceptable to me;

(5) All results will be treated with strict confidence, all individuals will remain anonymous in reporting any results, and all results will be handled in a professional manner;

(6) The University of Tennessee, its agents and employees, are released from any liability resulting from such participation, irrespective of cause or effect.

By my signature, I indicate that the research has been explained to me in detail and that I understand that any further questions that I may have about the project will be answered for me by the project director or some other designated member of the project staff.

Signed: _____

Witness: _____

Date: _____

DIETARY HISTORY

NAME_____EXPT. NO._____DATE_____

ADDRESS_____

BIRTH DATE_____

VEGETARIAN_____NON-VEGETARIAN_____NUMBER OF YEARS___

IF VEGETARIAN, DO YOU USE EGGS_____, DAIRY PRODUCTS_____, FISH_____

SINGLE_____ MARRIED_____NUMBER OF CHILDREN_____

ANY BROKEN BONES_____ AT WHAT AGE_____

MEDICATION_____

MEALS EATEN PER DAY: BREAKFAST_____ LUNCH_____SUPPER_____OTHER_____

IF "OTHER," EXPLAIN: _____

FOODS WELL LIKED AND EATEN OFTEN:

FOODS DISLIKED AND AVOIDED:

FOOD GROUPS—FREQUENCY OF SERVINGS

1. Bread and Cereals

   Bread: Whole grain_____ Enriched_____

   Cereals: Cooked_____ready-to-serve_____rice_____

   Number of servings per day_____

   Other: Pastas (macaroni, etc.)_____pancakes, waffles, doughnuts, sweet rolls_____

   Number of servings per week_____

2. Milk and dairy products

   Milk: whole_____ 2%_____ skim_____ buttermilk_____

   evaporated_____ dry non-fat (reconst.)_____

   Amount per day: 3 or more cups_____ 2-3 cups_____

   0-2 cups_____none_____

   Cheese: cottage_____cream_____cheddar type_____

   Number of servings per week_____

   Other: yogurt_____ice cream_____ice milk_____

   Number of servings per week_____

3. Fruits and vegetables

Citrus fruits (includes juice): oranges_____grapefruit_____
tangerines_____

Other juices: apple_____cranberry_____grape_____pineapple_____
prune_____

Number of servings per day_____

Other fruits: apples_____apricots_____bananas_____berries_____
grapes_____pears_____peaches_____

Number of servings per week_____

Vegetables: potato (white)_____ tomato, raw_____tomato, canned_____
green leafy, raw_____ green leafy, cooked_____
green, non-leafy, raw_____ green, non-leafy, cooked_____
deep yellow, raw_____ deep yellow, cooked_____ other_____

Servings per day_____

4. Meat and meat alternates

Meat: beef_____ veal_____lamb_____ pork_____ liver____fish____
poultry_____ luncheon meats_____ other_____

Number of servings per day_____

Alternates: eggs_____ dry beans_____ dry peas_____ lentils_____
nuts_____ peanuts_____ peanut butter_____ meat analogs_____

Number of servings per day_____

5. Miscellaneous

Fats and oils_____ butter or margarine_____ cookies_____ cake_____
molasses_____ syrup_____ candy_____ coffee_____ tea_____
cocoa_____ soft drinks_____ alcohol_____ tobacco_____

Frequency of use_____

NAME_____EXPT. NO._____

ADDRESS_____

DATE_____ DAY OF WEEK_____

| | FOOD | KIND & STATE | AMOUNT |
|---|---|---|---|
| BREAKFAST | | | |
| BETWEEN MEALS | | | |
| NOON MEAL | | | |
| BETWEEN MEALS | | | |
| EVENING MEAL | | | |
| AFTER EVENING MEAL | | | |

SUPPLEMENTS: VITAMIN____MINERAL____OTHER____BRAND_____

UNIVERSITY OF TENNESSEE
NUTRITION RESEARCH
INSTRUCTION SHEET FOR RECORDING FOOD INTAKE

We would like a record of what you eat for __7__ days.

Please read carefully the instructions below before you start to list the foods you have eaten.

Please record foods and snacks as they are eaten rather than trying to do a recall at the end of the day. If you need more space, use the back of the sheet.

1. WRITE DOWN EVERYTHING THAT YOU EAT

   If you miss a meal, write "nothing" in the space for that meal.

2. BE SURE TO WRITE DOWN THE KIND OF FOOD YOU EAT (KIND)

   Example: Cereal—Oatmeal, shredded wheat, cornflakes, etc.
   Bread——Whole wheat, white, rye; also commercial or homemade
   Meat——Roast beef, hamburger, veal steak, pork chops, etc.
   Salad——Head lettuce, canned fruit, tuna, cottage cheese,etc.
   Milk——Whole, 2%, skim, canned, etc.

3. DESCRIBE SPECIFICALLY HOW EACH FOOD IS PREPARED (STATE)

   Example:
   Egg — fried, boiled, scrambled, etc.
   Meats — broiled, breaded, fried, baked, etc.
   Fruits and vegetables— fresh, frozen or canned
   Vegetables — creamed, buttered, mashed, baked, etc.

   If food is not cooked, but eaten raw, write "RAW"

4. WHEN DIFFERENT FOODS ARE COMBINED WRITE DOWN EACH FOOD INCLUDED AND THE AMOUNT OF EACH FOOD

   Example:

| Raw Salad | | Cheese Sandwich | |
|---|---|---|---|
| lettuce | 1 leaf | bread | 2 slices |
| tomato | 1 slice | cheddar cheese | 1 slice |
| cucumber | 2 slices | lettuce | 1 leaf |
| French dressing | 1 tablespoon | mayonnaise | 2 teaspoons |

5. WHEN YOU EAT OTHER COMBINATION FOODS, SUCH AS CASSEROLE DISHES, SOUPS, STEWS, PUDDINGS, ETC., WRITE DOWN THE INGREDIENTS IF HOMEMADE OR SIMPLY THE BRAND NAME IF A CONVENIENCE OR STORE-BOUGHT ITEM IS USED.

   Example: Soup—Campbell's Tomato

6. WRITE DOWN THE AMOUNT OF EACH FOOD YOU EAT. Use a standard measuring cup, teaspoon or tablespoon, and a ruler to "measure" your food. Write down how many level teaspoons (t), tablespoons (T) you eat or whether you eat 1/2 or 1/3 or 1 cup, etc. Write down the number of

slices or pieces. For Example: pineapple, canned, 1 slice or apple, raw, 1 whole. Do not write down "glasses," "bowls," or "plates" for any foods such as milk, soup, vegetables, etc. Use the utensils provided to determine the amount.

Example: Soup—Campbell's Tomato 1 cup

The ruler should be used for foods that cannot be measured with a measuring cup, teaspoon or tablespoon. Some examples are cake, meat, pancakes, pies, etc. For foods with a round shape such as rolls, pancakes, meat patties, cupcakes, etc., the diameter and thickness should be measured. For all other shapes, length, width and thickness should be measured.

Example: pancake —1-8" diameter, 1/4" thick
choc. cake—iced, 1 piece, 2" x 3" x 1"
baked ham——1 slice, 4" x 3" x 1/4"
pie ——give measurements in inches, or tell whether it is a 1/4th or 1/8th etc. of a 8", 9" or 10" pie (diameter of whole pie)

7. BE SURE TO WRITE DOWN THE FOODS YOU ADD TO OTHER FOODS AND THE AMOUNT SUCH AS THE SUGAR, CREAM, OR BUTTER YOU USE.

   Example: the amount of sugar or cream used on cereal, fruit or in tea and coffee; the amount of butter on vegetables or bread; the amount of jelly on toast or syrup on pancakes

   Remember to record in level teaspoons or tablespoons; then if you want more, take it, just remember to add that amount, too.

   SAMPLE RECORDINGS:

| FOOD | KIND AND STATE | AMOUNT |
|---|---|---|
| cereal | oatmeal | 3/4 cup |
| sugar | - - - | 2 teaspoons |
| cream | half and half | 1/4 cup |
| pancake | Hungry Jack Pancake Mix | 1, 6" diam. 1/4" thick |
| egg | fried | 1 large |
| meat | baked ham | 4" x 2" x 1" |
| potatoes | mashed | 3/4 cup |
| peas | canned | 1/2 cup |
| butter on peas | - - - | 1/2 teaspoon |
| milk | whole | 1 cup |
| cake | choc., iced | 2" x 2" x 1" |

8. LIST AMOUNT AND BRAND OF ANY VITAMIN/MINERAL SUPPLEMENTS YOU TAKE.

9. IF YOU HAVE QUESTIONS, PLEASE DO NOT HESITATE TO CALL MRS. MASON OR DR. BEAUCHENE AT 974-3491.

## Instructions for Collection of a 24-hour Urine Specimen

Step 1 Void urine, discard it and record time.
Time_____.

Step 2 Collect all urine excreted during the next 24 hours—day and night—and pour it into the large container provided. If foil pans are used, rinse with water and allow to dry.

Step 3 Keep the plastic container cool—you do not need to refrigerate the sample, just keep it away from sources of heat.

Step 4 Exactly 24 hours after the first voiding (the one which was discarded in Step 1), void urine completely, save it, and add it to the large container.

Step 5 The large container now represents a 24-hour urine specimen. Members of the research team will pick up the containers.

# APPENDIX B

TABLE IX

PHYSICAL MEASUREMENTS[1] OF OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Parameter | Vegetarians | | | Nonvegetarians[2] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = | Lactoovovegetarians[3] (n = 57) | (n = 62) |
| Age (years) | 57.4 ± 1.4[a4] | 64.8 ± 5.2[b] | 55.9 ± 1.4[a] | 61.5 ± 1.3[b] |
| Height (cm) | 161.8 ± 0.8[a] | 161.2 ± 2.9[a] | 162.2 ± 0.8[a] | 161.6 ± 0.8[a] |
| Weight (kg) | 63.2 ± 1.6[a] | 49.5 ± 3.1[b] | 65.7 ± 1.8[a] | 66.6 ± 1.5[a] |
| Triceps Skinfold Thickness (mm) | 27.2 ± 1.0[a] | 19.1 ± 2.7[b] | 28.6 ± 1.1[a] | 32.0 ± 1.0[c] |
| Body Fat (%)[5] | 22.7 ± 1.1[a] | 13.9 ± 2.9[b] | 24.2 ± 1.2[a] | 27.7 ± 1.1[c] |
| Lean Body Mass (kg)[6] | 43.0 ± 0.6[a] | 38.8 ± 1.8[b] | 44.0 ± 0.7[a] | 43.1 ± 0.6[a] |

[1]Mean ± SEM.

[2]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[3]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[4]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

[5]Calculated using the formula: percent body fat = (triceps skinfold thickness - 6.16)/0.934.

[6]Calculated using the formula: LBM = 0.29569 (weight) + 0.41813 (height) - 43.2933.

TABLE X

MEAN DAILY ENERGY AND PROTEIN INTAKES[1] OF OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Nutrient | Vegetarians | | | Nonvegetarians[2] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[3] (n = 57) | (n = 62) |
| Energy (kcal/day) | 1516 ± 43[a4] | 1522 ± 100[a] | 1515 ± 45[a] | 1506 ± 43[a] |
| Total Protein (g/day) | 54.0 ± 1.3[a] | 50.8 ± 3.6[a] | 54.7 ± 1.3[a] | 65.9 ± 1.8[b] |
| Animal Protein (g/day) | 15.0 ± 1.5[a] | 3.1 ± 2.3[b5] | 16.7 ± 1.6[a] | 45.5 ± 1.8[c] |
| Vegetable Protein (g/day) | 29.2 ± 1.3[a] | 38.2 ± 2.4[b] | 28.2 ± 1.4[a] | 10.8 ± 0.7[c] |
| Mixed Protein (g/day)[6] | 10.7 ± 0.5[a] | 9.5 ± 0.8[a5] | 10.8 ± 0.6[a] | 10.2 ± 0.6[a] |
| Energy from Protein (%) | 14.5 ± 0.3[a] | 13.3 ± 0.4[b] | 14.7 ± 0.3[a] | 17.8 ± 0.4[c] |
| Protein Sources | | | | |
| Animal (% of total) | 27.4 ± 2.5[a] | 5.2 ± 3.5[b5] | 30.0 ± 2.7[a] | 68.0 ± 1.6[c] |
| Vegetable (% of total) | 54.0 ± 2.1[a] | 75.5 ± 2.4[b] | 51.7 ± 2.2[a] | 16.6 ± 1.1[c] |
| Mixed (% of total)[6] | 19.9 ± 0.4[a] | 19.3 ± 2.2[a5] | 19.7 ± 1.0[a] | 15.7 ± 1.0[b] |

[1]Mean ± SEM.

[2]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[3]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[4]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

[5]Appearance of consumption of animal protein by the vegans is due to calculation errors.

[6]Includes foods containing both animal and vegetable proteins.

TABLE XI

MEAN DAILY EXCRETION[1] OF URINARY COMPONENTS BY OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Metabolite | Vegetarians | | | Nonvegetarians[2] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[3] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 8.27 ± 0.32[a4] | 8.34 ± 0.85[a] | 8.29 ± 0.35[a] | 8.92 ± 0.37[a] |
| Urea Nitrogen (g/day) | 7.00 ± 0.32[a] | 7.01 ± 0.74[a] | 6.97 ± 0.34[a] | 7.72 ± 0.38[a] |
| Ammonia Nitrogen (mg/day) | 170.8 ± 12.4[a] | 193.9 ± 16.3[ab] | 168.7 ± 13.1[a] | 226.2 ± 17.0[b] |
| Creatinine (g/day) | 1.32 ± 0.05[a] | 1.21 ± 0.12[a] | 1.34 ± 0.04[a] | 1.53 ± 0.06[b] |
| Hydroxyproline (mg/day) | 32.3 ± 1.5[ab] | 28.0 ± 1.7[a] | 33.2 ± 1.6[b] | 35.3 ± 1.8[b] |
| Inorganic Sulfate (g/day) | 1.50 ± 0.07[a] | 1.35 ± 0.17[a] | 1.55 ± 0.07[a] | 1.56 ± 0.08[a] |
| Sources of Nitrogen | | | | |
| Urea (% of total) | 84.3 ± 1.6[a] | 84.1 ± 2.3[a] | 85.5 ± 1.7[a] | 86.2 ± 1.5[a] |
| Ammonia (% of total) | 2.12 ± 0.14[a] | 2.47 ± 0.36[ab] | 2.11 ± 0.15[a] | 2.64 ± 0.17[b] |
| Creatinine (% of total) | 6.50 ± 0.33[a] | 5.57 ± 0.40[b] | 6.53 ± 0.35[a] | 6.95 ± 0.30[a] |

[1]Mean ± SEM.

[2]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[3]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[4]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

TABLE XII

RATIOS OF URINARY METABOLITES[1] IN OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Ratio | Vegetarians | | | Nonvegetarians[2] |
|---|---|---|---|---|
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[3] (n = 57) | (n = 62) |
| Inorganic Sulfate/ Total Nitrogen | 0.18 ± 0.01[a4] | 0.16 ± 0.01[a] | 0.18 ± 0.01[a] | 0.18 ± 0.01[a] |
| Inorganic Sulfate/ Urea Nitrogen | 0.22 ± 0.01[a] | 0.19 ± 0.01[b] | 0.22 ± 0.01[a] | 0.21 ± 0.01[a] |
| Inorganic Sulfate/ Creatinine | 1.16 ± 0.05[a] | 1.13 ± 0.10[a] | 1.19 ± 0.05[a] | 1.05 ± 0.05[a] |

[1]Mean ± SEM.

[2]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[3]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

[4]Means in a row not sharing a common superscript are significantly different ($P < 0.05$).

# APPENDIX C

TABLE XIII

CORRELATION OF TOTAL PROTEIN INTAKE WITH URINARY METABOLITES AND RATIOS OF THOSE METABOLITES IN OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Parameter | Correlation Coefficient | | | |
|---|---|---|---|---|
| | Vegetarians | | | Nonvegetarians[1] |
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[2] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 0.31** | 0.83* | 0.31** | 0.20 |
| Urea Nitrogen (g/day) | 0.28* | 0.90** | 0.25* | 0.20 |
| Ammonia Nitrogen (mg/day) | 0.06 | -0.28 | 0.13 | -0.04 |
| Creatinine (g/day) | 0.28* | 0.40 | 0.27* | 0.19 |
| Hydroxyproline (mg/day) | 0.25* | 0.67 | 0.26* | -0.09 |
| Inorganic Sulfate (g/day) | 0.23 | 0.90** | 0.19 | 0.32** |
| Inorganic Sulfate/ Total Nitrogen | -0.06 | 0.49 | -0.12 | 0.26* |
| Inorganic Sulfate/ Urea Nitrogen | -0.04 | 0.46 | -0.04 | 0.18 |
| Inorganic Sulfate/Creatinine | -0.01 | 0.71 | -0.03 | 0.18 |

[1]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[2]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

*P < 0.05.

**P < 0.01.

TABLE XIV

CORRELATION OF ANIMAL PROTEIN INTAKE WITH URINARY METABOLITES AND RATIOS OF THOSE METABOLITES IN OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| | Correlation Coefficient | | | |
|---|---|---|---|---|
| | Vegetarians | | | Nonvegetarians[1] |
| Parameter | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[2] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 0.07 | | 0.14 | 0.30* |
| Urea Nitrogen (g/day) | 0.07 | | 0.14 | 0.30* |
| Ammonia Nitrogen (mg/day) | -0.07 | | -0.06 | -0.01 |
| Creatinine (g/day) | 0.41*** | | 0.44*** | 0.33** |
| Hydroxyproline (mg/day) | 0.31** | | 0.31** | 0.10 |
| Inorganic Sulfate (g/day) | 0.30** | | 0.30** | 0.31** |
| Inorganic Sulfate/ Total Nitrogen | 0.46** | | 0.38** | 0.11 |
| Inorganic Sulfate/ Urea Nitrogen | 0.56*** | | 0.51*** | 0.07 |
| Inorganic Sulfate/Creatinine | 0.03 | | 0.01 | 0.06 |

[1]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[2]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

*P < 0.05.

**P < 0.01.

***P < 0.001.

TABLE XV

CORRELATION OF VEGETABLE PROTEIN INTAKE WITH URINARY METABOLITES AND RATIOS OF THOSE METABOLITES IN OLDER ADULT FEMALE VEGANS, LACTOOVOVEGETARIANS AND NONVEGETARIANS

| Parameter | Correlation Coefficient | | | |
|---|---|---|---|---|
| | Vegetarians | | | Nonvegetarians[1] |
| | All (n = 63) | Vegans (n = 6) | Lactoovovegetarians[2] (n = 57) | (n = 62) |
| Total Nitrogen (g/day) | 0.02 | 0.59 | -0.11 | -0.05 |
| Urea Nitrogen (g/day) | -0.01 | 0.65 | -0.15 | -0.05 |
| Ammonia Nitrogen (mg/day) | 0.08 | -0.52 | 0.11 | 0.09 |
| Creatinine (g/day) | -0.05 | -0.02 | -0.14 | -0.09 |
| Hydroxyproline (mg/day) | -0.03 | 0.60 | -0.05 | -0.01 |
| Inorganic Sulfate (g/day) | -0.13 | 0.67 | -0.21 | 0.06 |
| Inorganic Sulfate/ Total Nitrogen | -0.21 | 0.43 | -0.18 | 0.19 |
| Inorganic Sulfate/ Urea Nitrogen | -0.15 | 0.39 | -0.06 | 0.15 |
| Inorganic Sulfate/Creatinine | -0.12 | 0.87* | -0.15 | 0.17 |

[1]Includes 1 subject who had 4 servings of meat and 2 who had 6 servings of meat each per week.

[2]Includes 4 subjects who had 1 serving of meat each and 2 who had 2 servings each per week.

*$P < 0.05$.

# VITA

Mary Elizabeth Kunkel was born in Newport, Arkansas, on September 8, 1953, where she attended public schools and was graduated from high school in May 1971. In the fall of that year, she entered the University of Central Arkansas and was graduated with a Bachelor of Science in Education with a major in home economics in May 1975. In August 1976, she received a Master of Science with a major in nutrition from The University of Tennessee, Knoxville. In August 1979, she received a Doctor of Philosophy in home economics from The University of Tennessee, Knoxville. While at The University of Tennessee, she served for 1 year as a graduate assistant and for 2 years as a graduate teaching assistant in the Department of Food Science, Nutrition and Food Systems Administration. Upon completion of the Ph.D., she accepted a postdoctoral training grant with the Institute of Dental Research, University of Alabama, Birmingham.