ABSTRACT OF THESIS

FACTORS OF NUTRITIONAL SIGNIFICANCE IN THE BLOOD OF MEN IN THE SEVENTH DECADE OF LIFE

Submitted by

Betty Jane Allyn Hunt

In partial fulfillment of the requirements

for the Degree of Master of Science

Colorado

Agricultural and Mechanical College

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ABSTRACT

There is an increase in the percentage of the population over 65 years of age. This proportionate increase creates the problem of maintaining the health and usefulness of the older segment of the population. It is thought that much can be done to maintain the health of the aged by adequate nutrition. Since little is known of the nutritional status of the aged, it is first necessary to set up standards by which to evaluate the nutritional state of this group.

The use of blood analysis to determine nutritional status is coming into more and more use. There has been started in the west, a large regional nutritional study in which newer methods of analysis adapted for small amounts of blood are being used. Since nutritional investigations both with these methods and among the aged are few, a study combining the two seemed of value. Too, no study of this nature among aged subjects at high altitudes was found in the literature.

Because of the foregoing reasons, i.e., an increase in the percentage of older people, the need for nutritional studies and the establishment of blood values at 5000 feet for older people, and the importance of testing newer methods on this age group, this study was initiated.

The problem

What are the values of selected factors of nutritional significance in the blood of a representative group

of aged people living in the same community?

Problem analysis.--1. What are the values for ascorbic acid, hemoglobin, plasma protein, hematocrit and erythrocyte count for this group as determined on fingertip blood?

2. Are these methods of blood analysis, adapted for use on small amounts of blood, feasible for nutrition field studies among this age group?

3. How do these values compare with published values for the same age group?

4. How do the values compare with published values for different age groups?

5. Do values show any trends with age?

6. What factors explain any differences or similarities found?

7. What is the possible effect of food habits?

Delimitation.--This investigation was limited to 25 men between the ages of 70 and 80 living independently at 5000 feet altitude.

The subjects lived independently of other families, relatives or institutions. According to the health histories they reported, all men were in good health and considered themselves well and active for their ages.

All subjects kept a record of their diets the day before the first blood sample was taken. Other information

about their food habits was secured. The diets were classified into Grades 1, 2 and 3, or good, fair and poor respectively. A commonly used standard (Basic 7 foods) was used as the guide for classification.

Fingertip blood samples were analyzed for hemoglobin, erythrocyte count, hematocrit, plasma ascorbic acid and specific gravity of the plasma. The samples were collected twice, at an interval of one week, at the same time of day.

An alkaline hematin method for the determination of hemoglobin was employed. Samples of blood for the erythrocyte counts were diluted with Hayem's solution and the counts were made on an improved counting chamber. Hematocrit was determined by a micro-method. Small bore capillary tubes, 1.5-2.0 by 100 millimeters, were partially filled with blood directly from the finger leaving generous space at either end for sealing with plicene cement. The tubes were centrifuged at 2700 revolutions per minute for 45 minutes. The amount of packed red cells were measured on a centimeter ruler. The values for hematocrit were expressed in percent.

The ascorbic acid of the plasma was determined by the Farmer and Abt micro-method.

The specific gravity of the plasma, used as an index of the plasma protein, was determined by Phillip's copper sulfate method.

The mean values for the above factors of the blood as determined by the indicated methods were: Red cell count

5.556 million cells per cubic millimeter of blood; hematocrit, 49.0 percent; ascorbic acid, 0.61 milligrams per 100 milliliters plasma; and plasma specific gravity, 1.0212. The hemoglobin values were not averaged because of difficulty with the method. The "typical" diets by qualitative comparison to the "Basic 7" showed that all except seven of the subjects had ostensibly inadequate diets.

The mean values for erythrocyte count and hematocrit, were higher than those for young or aged subjects at lower altitudes. Altitude was probably one of the primary factors accounting for the higher values. The hemoglobin values were presumably within the normal range. The mean values for ascorbic acid of the plasma were comparable to, although somewhat lower than, the values for young adults as found in other surveys. However, the mean ascorbic acid value was lower than that of aged subjects on a controlled good institutional diet. The study showed that the subjects who did not have citrus fruit or tomatoes in their "typical" diet tended to have lower plasma ascorbic acid values. The mean value for the specific gravity (indicating protein) of the plasma was below the normal range for adults. Although diets were poor, they appeared to be adequate in protein. There were no known serious diseases which would account for the lower plasma protein values.

The dietary items in which the majority of subjects

were deficient were milk and enriched fat, and next in order of deficiency were fruits and vegetables. In many of the diets, fruits and vegetables were eaten often enough but not in large enough quantities to give the recommended allowances. Most of the subject's diets showed that milk and enriched fats simply were not used.

This study indicated that mass nutritional study of aged subjects needs more careful planning and more time than similar studies among younger subjects. Methods using fingertip blood were found to be feasible for this age group. However, further study is needed on the hematocrit and hemoglobin methods.

This study indicated the need for further nutritional investigations of large numbers of older subjects at higher altitudes in order to establish standard values.

T H E S I S

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COLORADO AGRICULTURAL AND MECHANICAL COLLEGE

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I HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER MY SUPERVISION BY BETTY JANE ALLYN HUNT

ENTITLED FACTORS OF NUTRITIONAL SIGNIFICANCE IN THE BLOOD OF MEN IN THE SEVENTH DECADE OF LIFE.

BE ACCEPTED AS FULFILLING THIS PART OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE

MAJORING IN FOODS AND NUTRITION

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Chapter I

INTRODUCTION

Much stress has been laid upon studies of nutritional status during the past few years. The National Research Council (22) has become concerned over the widespread dietary inadequacies revealed by these surveys. Most of the studies have been conducted among young adults with comparatively few among aged people.

However, aged people constitute a big problem in this country. There is a population shift toward people aged 65 years. The United States Census for 1940 (43:2) showed that there were slightly more than nine million people over the age of 65, or 6.9 percent of the total population, an increase of 33 1/3 percent since 1900. Stieglitz (39:13-14) implied that this situation created the problem of maintaining the health and usefulness of these new millions.

The blood values for a few well chosen items may show a somewhat general nutritional picture. They may reflect the value to the body of the many ingested nutrients which are furnished by individual foods we consume.

Nutritional studies utilizing blood values are less subject to personal subjective error and are less time consuming than detailed dietary studies which involve weighed diets and their chemical analysis. They also obviate much training and effort on the part of the subject. The latter are often reasons for failing to obtain cooperation and for failing to get accurate results in nutritional studies.

Among factors which are commonly measured in blood samples for nutritional status studies are ascorbic acid, hemoglobin, plasma protein, hematocrit and to a lesser extent, erythrocyte count. Ascorbic acid is widely used for the determination of the quality of some foods, and it is assumed that the ascorbic acid value of the blood is an indication of the quality of these foods eaten and the value of them to the individual's body. Since anemia may result from various nutritional deficiencies, blood values for hemoglobin, hematocrit and red blood cell count, if abnormal, may denote some nutritional deficiencies and/or pathological conditions. It is commonly accepted by medical workers that hemoglobin and red blood cell counts are higher at higher altitudes. (Todd and Sanford, 41:217; Miller, 19:1174; Lurie, 17). However, information on blood values of supposedly normal older subjects at 5000 feet is needed. The protein content of the blood is of significance

because it reflects, to some extent, the protein utilization of the body.

Colorado is cooperating on a regional nutritional study which was recently started in Oregon. Methods adapted to the use of small quantities of fingertip blood were used. Since no other state in the region has started a study of older people, it will be of value to determine the workability of these methods and the reactions of older people to this type of nutritional study.

Because of the foregoing reasons, i.e., an increase in the number of older people, the need for nutritional studies and the establishment of blood values at 5000 feet for older people, and the importance of testing newer methods on this age group, this study was initiated.

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Delimitation.--This investigation was limited to 25 men between the ages of 70 and 80 living independently at 5000 feet altitude.

Chapter II

REVIEW OF LITERATURE

Numerous surveys of nutritional status using blood values as indices have been made. Almost all of these surveys have been conducted on young adults. A few pertinent studies are reviewed below:

Several large-scale state and regional nutritional studies of college students have been made. Brown et al. (2) in the western region made a survey of the ascorbic acid nutrition of college students. The mean plasma values for men were found to be significantly lower than those of women. Fifty percent of the women and 76 percent of the men had values below 0.8 milligrams of ascorbic acid per 100 milliliters of plasma. This value, 0.8 milligrams, was considered "normal". Ohlson et al. (24) reported the blood values for a survey of college women of the North Central region. They concluded that the normal values for hemoglobin, erythrocyte counts, and hematocrit must be interpreted as a range wider than those recorded by earlier studies. Sheets and Barrentine (36) of Mississippi surveyed college men and women. The average values for hemoglobin were lower than those reported by other

investigators. The erythrocyte counts were within normal limits, but were also low. Dodds and MacLeod (5) reported a study of ascorbic acid nutrition among Tennessee college women. They found that the mean values and distribution of the values for ascorbic acid agreed well with other surveys on similar population groups. In Alabama, McMillan and Todhunter (18) found that 37 percent of the college women they surveyed had satisfactory values for both nutritional factors, hemoglobin and ascorbic acid. They found that there was no correlation between the hemoglobin and plasma ascorbic acid values of the same individual, i.e. that one did not affect the other. Donelson et al. (8) reported the total findings of the midwestern survey. (The hematological phase of this survey was reported earlier by Ohlson et al., 24). They concluded that the mean hematocrit, hemoglobin, and erythrocyte counts were within normal limits, but that the mean ascorbic acid values were low for the group of college women studied.

High altitude causes some change from normal blood values at sea level. Lurie (17) conducted some research at 5740 feet altitude in South Africa with 30 people of each sex. He reported that hemoglobin, erythrocyte counts and cell volume were proportionally higher at that altitude, as compared with normal data

at sea level.

A relatively small number of blood studies, nutritional or otherwise, have been conducted with elderly subjects.

An outstanding series of studies has been carried on among this age group by Rafsky and Newman (28-34). However, even though the subjects were considered "normal", they were confined to a home for the aged. They (33) determined the blood levels of ascorbic acid and conducted a saturation experiment with 25 aged subjects 66 to 83 years. They found that the overall average for plasma ascorbic acid (0.72 milligrams percent) before dosage was somewhat below the range considered normal for young adults. Newman and Gitlow (23) used subjects from the same type of institution for hematological study of 50 aged males and females. Hemoglobin, erythrocyte count, packed cell volume and corpuscular constant values tended to be lower than those found by other investigators of younger subjects. Fowler et al. (11) conducted a hematological study among 100 elderly hospital patients free of any serious debilitating diseases. Seventy-three subjects were men and 27 women, of which the overall average age was 71.3 years. Their study showed a slight reduction from normally accepted values for young adults in hemoglobin, erythrocyte and hematocrit. Miller (19) made a study of 160 men between

the ages of 60 and 104 years. The subjects were from the New York City Farm Colony (presumably for the aged). He found that the hemoglobin and red blood cell count decreased in the aged; and that the decrease in hemoglobin was proportional to the decrease in red blood cells. Stieglitz (39:818) reported the values for 156 healthy men 50 years and older. The average values for erythrocytes (4.60 million) and hemoglobin (14.22 grams) were lower than the values for younger people, but still within the normal range.

Bürker (3) reported that the total hemoglobin increases in old age; also that the values for women tended to approach the values for men. He attributed these changes to more difficult breathing, changes in heat regulation, and in women, cessation of the sex cycle.

The reliability of the methods and the interpretation of experimental data in studies of the above type are constantly being reviewed in an effort to arrive at more normal standards. Concerning ascorbic acid, opinion differs as to the significance of the plasma values. Todhunter and Robbins (42) stated that plasma ascorbic acid was possibly a more reliable index of ascorbic acid nutrition of an individual than urinary excretion because it was not subject to so many daily variation. Smith et al. (38) also found that daily

variations in intake were not apparent unless they were of considerable magnitude. Dodds and MacLeod (7) found that ascorbic acid plasma values gave little indication of the present intake of the vitamin. In a later study by these same authors (6), they confirmed this theory and stated that the intake of ascorbic acid cannot be narrowly defined by the plasma values. Johnson et al. (15) stated that blood ascorbic acid values give comparable results regardless of the time of day the sample is taken. Evidence by McMillan and Todhunter (18) showed that there is less than 0.20 milligrams average variation of the plasma value of any one person on non-consecutive day sampling. They also pointed out that there is no generally accepted standard for hemoglobin or ascorbic acid. Haines et al. (14) indicated that plasma levels of ascorbic acid reflect the stores and intake over a period of time rather than the immediate intake.

In view of the many surveys of supposedly normal people that reveal low blood values for ascorbic acid, Munsell et al. (20) made a noteworthy comment: that low levels of ascorbic acid in the blood for those used to a low ascorbic acid diet is not indicative that they should have scurvy.

In spite of the controversy over interpretation of data, ascorbic acid values continue to be used and

have yielded valuable information about ascorbic acid nutrition.

The determination of protein values in blood plasma or serum are commonly used in studies as an index of protein nutrition of the body. Hypoproteinemia is found in a number of abnormal conditions. Muntwyler (21) stated that hypoproteinemia is encountered in liver, kidney, and bone diseases, in chronic infections and in malnutrition. Frisch (12) showed that nutritional edema in rats was associated with markedly reduced levels of serum protein. Graafland (13) found that hypoproteinemia resulted from a low calorie diet even though sufficient animal protein was given. His hypothesis was that the protein was used for calories.

A number of methods for determining the amount of protein in the blood have been perfected. Many of these methods are long, tedious and impractical for survey work. The use of specific gravity to determine blood protein is one of the more simple. Adams and Ballou (1) attested to the reliability of this method by stating that the specific gravity method of determining plasma and serum protein was comparable to the micro-Kjeldahl method.

Hence, it has been found that blood studies of independent aged subjects have been scanty and inadequate. The studies among aged subjects showed that values for

plasma ascorbic acid, erythrocyte count, hematocrit, and hemoglobin have tended to be lower, with one exception, than those values accepted as normal for young adults.

Chapter III

MATERIALS AND METHODS

In order to ascertain the values for the blood of aged individuals the following materials and methods were used.

Subjects

Twenty-five active men between the ages of 70 and 80 were used in the experiment. The subjects were selected upon the bases of age, health, degree of activity and willingness to participate in the experiment. All subjects were active enough to take care of themselves. None were mentally deficient or dependent upon others for their personal care. All subjects lived independently, i.e. were not living with other families, relatives or in an institution. They were all Fort Collins residents.

Prospective subjects were called upon to explain the experiment and to obtain their cooperation in the study. If they agreed to participate a second visit was made. At this time, the health history was obtained, the food record was explained and appointment times for the blood collection were made.

Food record

A one day diet record was kept by the subjects the day before the first blood sample was taken. The subjects were given instructions to "write down everything you eat during that day; everything you put in your mouth and swallow". Later, the author went over the menu with the subject and filled in any omitted details. The menus were copied on food record sheets (see Appendix A) and summarized together with other information (Appendix B). All amounts of the recorded food were suggested by the subject or were the author's interpretation of amounts from graphic description. These amounts were then translated into terms of servings. Each diet was evaluated in terms of servings and classified as Grade 1, 2 or 3 according to the following plan.

	Grade 1* (Minimum servings)	Grade 2 (Minimum servings)	Grade 3
Whole grain or enriched cereal products.	2	2	(One serving less of anything in a Grade 2 diet)
Vegetables and/or fruits	4	2	
Citrus fruit or tomatoes	1		
Meat or protein substitute	1 — either		
Eggs per week	3 /		
Milk	2	1	
Fat (enriched)	2 (T)	1 (t)	

* One serving less of any one constituent in a Grade 1 diet was allowed for daily variation.

The diet classifications, Grade 1, 2, and 3, denoted the quality of the diets of the subjects. A

Grade 3 diet is obviously inadequate. A Grade 2 diet, at the minimum, is not adequate but still not poor. A Grade 1 diet would probably meet minimum adequate standards. Detailed analysis was not justified because only one day food records were feasible in this study.

Health histories

Health histories (see Appendix C and D) were filled in by the interviewer and were not presumed to be a medical examination or detailed medical records. Data were recorded as much as possible in terms expressed by the subject. Although the guide sheet contained technical terms as a space-saver, the questions were asked in laymen's terms such as, "Have you ever had any kidney trouble?" The subjects were cooperative and the data were considered accurate accounts of the subject's knowledge of his health history. The subject's word was taken for his age, height and weight.

The records were modeled after those used in a regional study of nutritional status being conducted currently in the western states and were modified for this age group.

Blood collection

Each subject was brought to the laboratory for blood collection in the morning after a light breakfast. Each subject gave two samples of blood exactly one week apart, except in the cases of two subjects; one was unable to come on the exact day because of his work, and the other

gave only one sample. The blood samples were taken very close to the same time each day and all were within one and a half hours variation.

There were several reasons for taking two samples. A large number of subjects could not be used because of the excessive time required to locate subjects of the proper age. A great deal of time was required to contact and interview each new subject. A second blood collection could be taken from the same subject without repeating the interviews. Two blood samples were thought to be worthwhile as a check on the values determined and as an indication of the variability of the values.

The blood was taken from the fingertip after the hand had been warmed well with hot water. A registered technician pricked the finger with a Bard-Parker blade and assisted with blood collection. As much free flowing blood as possible was obtained and if that wasn't enough, the finger was squeezed gently.

Methods of analysis of blood (In order of collection)

Hematocrit.--Small bore capillary tubes (melting point tubes), 1.5-2.0 millimeters in diameter and four inches long, were dipped in about one half inch deep finely ground potassium and ammonium oxalate in the ratio of 2 to 3 by weight, respectively. The excess oxalate was tapped out of the tubes. Tubes were partially filled with blood directly from the finger,

leaving about one half inch on either end for sealing. The tubes were laid aside until after all the rest of the blood had been collected. They were then sealed with plicene cement and labeled with a small adhesive tape with the subject's number. The samples were centrifuged for forty-five minutes at 2700 revolutions per minute. Relative volumes of packed cells and plasma were measured with a centimeter ruler, using the same ruler for all samples. Readings were taken to the nearest tenth of a centimeter. Values were expressed in percent.

Ascorbic acid.--Fingertip blood was collected in small blood vials about one fourth inch in diameter and one half inch in length. Two drops of two percent potassium and ammonium oxalate solution (ratio 2 to 3) were dried previously in each tube. They were measured into the vials by means of a roughly calibrated 5 cubic millimeter constricted pipette. One drop was equal to approximately 5 cubic millimeters of oxalate solution. The tubes were filled one fourth to one third full of blood with special care taken to prevent clotting during collection. The samples were corked immediately and refrigerated at all times except when being handled.

They were analyzed for reduced ascorbic acid by the Farmer and Abt micro-method (9) within six hours

after collection. One determination per sample was made. The dye used in the determination was diluted daily from stock solution that was made fresh weekly. The diluted dye was standardized daily against pure ascorbic acid solution that was made fresh every 48 hours. Metaphosphoric acid solution was made fresh weekly. The ascorbic acid solution, dye, and acid used were refrigerated except when in use. Values for ascorbic acid were expressed as milligrams per 100 milliliters plasma. The accuracy of the method was checked by a recovery test. The recovery tests averaged 102 percent recovery of ascorbic acid which was within the limits of experimental error.

Erythrocyte count and hemoglobin.--Blood samples for these two determinations were collected last and were interchangeable in order according to the size of the drop of blood on the end of the finger. Most samples were taken directly from the finger. If the subject bled profusely and the blood did not clot too rapidly, the blood was allowed to drop into a spot plate from which it was measured. The blood samples for erythrocyte determinations were diluted with Hayem's solution and counted on an improved counting chamber, (Todd and Sanford, 41:216-228). Once a week, one chamber was recounted in a different order to check the accuracy of the count. The counting was found to be accurate.

Hemoglobin was determined by a combination of alkaline hematin methods (Storvick, 40, and Coleman, 4). A 20 cubic millimeter marked Adams pipette was used. Four milliliters of one percent ammonium hydroxide was the diluting solution. The optical density of the samples were read on a Coleman spectrophotometer set at a wave length of 542 according to the null method. Samples were corked, but not refrigerated, until read. Samples were read within one hour after collection.

The pipettes used for hemoglobin measurement were checked for content and found to be within the limits of experimental error.

Plasma protein.--Determination of the specific gravity as a measure of the plasma protein was first attempted with a gradient tube according to the method of Lowry and Hunter (16). This proved unsuccessful so the copper sulfate method of Phillips (25) was used. The plasma used was either the excess from the ascorbic acid blood vial or the plasma from the hematocrit tubes. Since the amount of oxalate was too small to change the readings, correction was not made for oxalate. A pyknometer was used to check the specific gravity of the copper sulfate solutions.

Precautions used in blood collection

Special precautions always had to be followed to avoid difficulty with hemolysis. Preliminary work showed the necessity for the following measures:

The hematocrit tubes must be well tapped free of excess oxalate to prevent hemolysis. Also, they must not be filled too full or the heat from the plicene cement used in sealing will cause hemolysis. When collecting the blood in the vials, the blood collection should be interrupted at least once to rotate the tube so that the blood will become well oxalated throughout the tube. One drop deficiency or excess of oxalate solution caused trouble with either clotting or hemolysis respectively. Too strenuous squeezing of the finger to extract blood also caused hemolysis.

Chapter IV

RESULTS

The results of the analysis of the fingertip samples of blood from elderly male subjects, and observations from their diet records and health histories are presented below.

Blood studies

The mean values for erythrocyte count, specific gravity of the plasma, hematocrit and reduced ascorbic acid for each of the subjects for the two samplings are given in Table 1. The subject's age, height and weight are also recorded in this table. The individual values are tabulated in Appendix E. The means, ranges and standard deviations are recorded in Table 2. The distribution of the mean value for erythrocyte counts, plasma specific gravity and hematocrit are illustrated in Figure 1 according to the frequency in different intervals. All mean values are an average of two determinations, except where marked.

The ages ranged from 70 to 78 years with the mean age being 73.5 years. The mean value for erythrocyte counts was 5.556 million cells per cubic millimeter. Nine of the subjects had mean values between 4.80 and

Table 1.--NUMBER, DESCRIPTION AND MEAN BLOOD VALUES OF SUBJECTS

Subject number	Age years	Height (1)	Weight pounds	Erythrocytes million	Plasma (2)	Ascorbic acid (3)	Hematocrit percent
2	70	5-5	165	5.11	1.020°	1.22°	53
17	70	5-7	165	5.69	1.020°	0.16	49
9	70	5-10	185	5.52	1.021	0.74	49
30	71	5-9	155	5.79	1.022°	1.19°	50
25	71	5-6.5	174	4.99	1.020°	0.18°	45°
24	72	5-9	200	5.39	1.023°	0.84°	47
27	72	5-8	152	5.92	1.017°	0.43°	55
23	72	5-7	146	5.50	1.019°	1.00	50
22	72	5-1.5	120	6.17°	1.020°	0.58°	48
16	72	5-7	116	5.12	1.020	0.25	49
32	72	5-9.5	184	6.41	1.022	1.12	56
20	73	5-7	140	5.58	1.023°	--	48
13	73	5-9	150	5.23	1.019	1.33	46
14	73	5-4	130	6.21	1.020	0.82	44
4	74	5-5	120	6.17	1.023°	0.47°	47
29	74	5-9	170	5.21	1.020°	0.94	44
1	74	5-7	162	5.79	1.023	0.77	48
21	76	6-0	170	5.12	1.017°	0.20	48
7	76	5-10	155	4.80	1.021	0.52°	49
31	76	6-1	150	5.53	1.024	0.60	45
12	76	5-8	145	5.60	1.020	0.28	48
3	77	5-5.5	205	5.99	---	0.43°	59
28	77	5-7	120	5.28	1.023	0.40	49
5	78	5-10	180	5.14	1.024°	0.11	53°
6	78	5-10	162	5.64	1.028°	0.14	46

(1) Feet and inches
(2) Specific gravity
(3) Milligrams per 100 milliliters plasma
° Only one value

Table 2.--AVERAGE VALUES FOR CONSTITUENTS IN THE BLOOD OF AGED SUBJECTS.

Factor	Mean	Range	Standard Deviation
Erythrocyte count (millions)	5.556	4.80-6.41	0.426
Plasma specific gravity	1.0212	1.017-1.028	0.00122
Ascorbic acid (milligrams per 100 milliliters of plasma)	0.608	0.11-1.33	0.383
Hematocrit percent	49.0	44-59	3.72

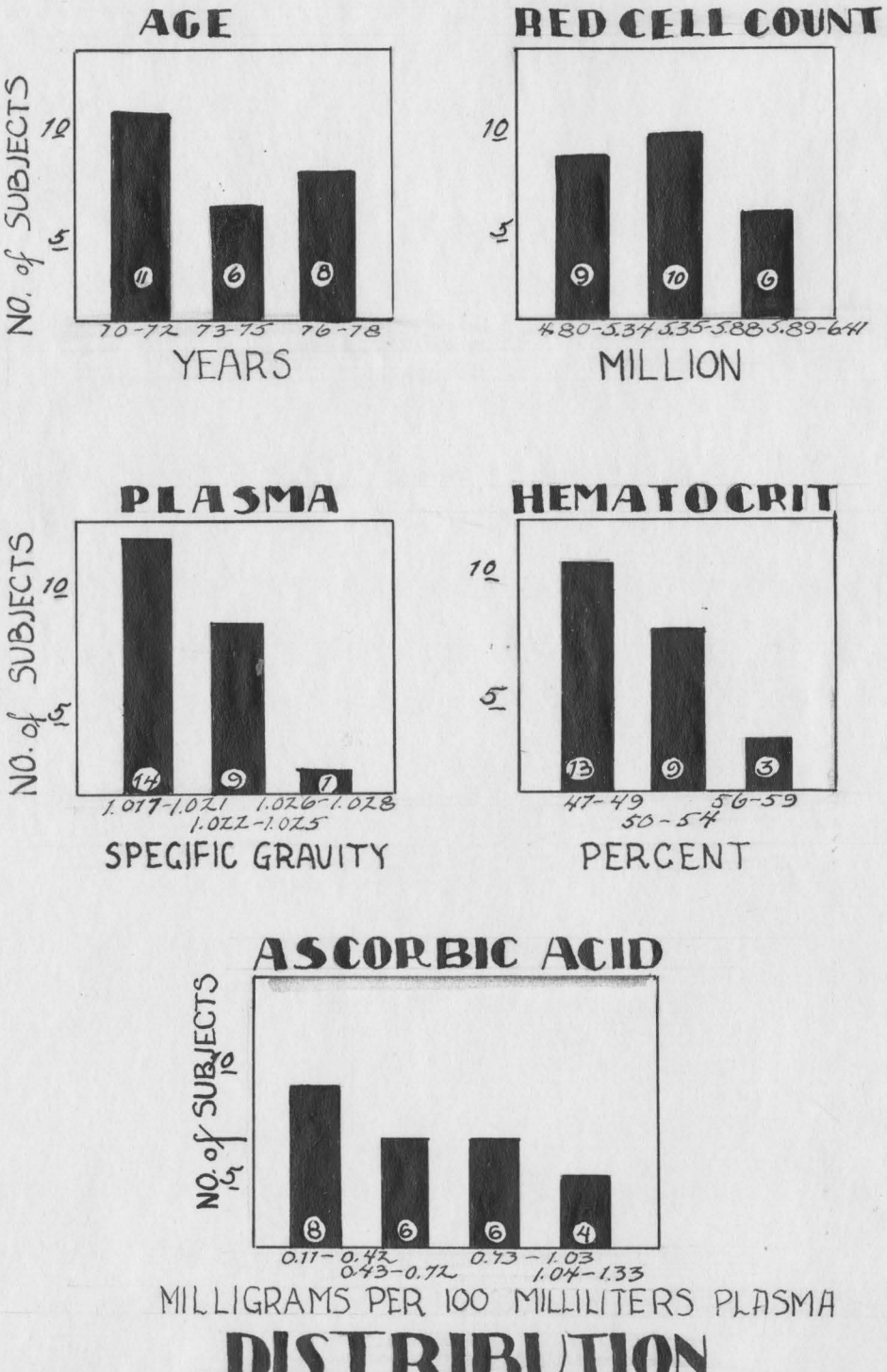

Figure 1.--DISTRIBUTION OF THE VALUES FOR CONSTITUENTS IN THE BLOOD

5.30 million per cubic millimeter; 10 between 5.31 and 5.90 million; and six between 5.91 and 6.45 million per cubic millimeter. The range was 4.80 to 6.41 millions, the standard deviation was 0.462 million.

Hemoglobin values are not given individually but are discussed in the following chapter.

The mean hematocrit value was found to be 49 percent. The range was 44 to 59 percent. Thirteen subjects had values which fell between 44 and 49; nine between 49 and 54; and three between 54 and 59 percent. The standard deviation was calculated to be 3.7 percent.

The mean value for specific gravity of the plasma was 1.0212 or 4.59 grams of protein per 100 milliliters of plasma. All except one value was below normal. The individual values clustered very closely around the mean. The range was 1.017 to 1.028 and the standard deviation 0.0012.

The mean value for reduced ascorbic acid was 0.61 milligrams per 100 milliliters of plasma. The mean values of the two samples for each subject ranged from 0.11 to 1.33 milligrams per 100 milliliters of plasma. Eight subjects had mean values between 0.11 and 0.42 milligrams; six between 0.43 and 0.72; six between 0.73 and 1.03; and four between 1.04 and 1.33 milligrams. The standard deviation was 0.383 milligrams per 100 grams.

Health records

All subjects except three were found to be in good health and quite active for their ages. Two of these had more limited activity because of "bad backs". One of these was under a doctor's care; the other was not. The third was still under a doctor's routine supervision after having a goiter removed six months ago. In spite of these things, the subjects considered themselves in fairly good health. Two of these subjects were on restricted diets advised by their physicians.

In all, general condition of the subjects was found to be good. The record of the past and present diseases showed that all were free from serious debilitating illnesses. Colds were the most frequent complaint and most of the subjects had had pneumonia sometime during their lives. Most cases of the latter occurred at the time of the first world war.

Food records

Two of the 25 subjects had Grade 1[1] diets and two subjects had Grade 2; the rest (21) had Grade 3 or poor diets. No subject's diet was deficient in cereal products. Fourteen subjects did not have any citrus fruit or tomatoes. The dietary items in which the

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1. For explanation of grades see page 19.

majority of subjects were deficient were milk and enriched fat; next in order of deficiency were fruits and vegetables. In many of the diets, fruits and vegetables were eaten often enough but not in large enough quantities to give the recommended allowances. Most of the subjects' diets showed that milk and enriched fats simply were not used.

Chapter V

DISCUSSION

On the whole, the mean values for the blood constituents determined in this study are not comparable to any one other study. The mean erythrocyte value (5.556 million cells) was most comparable to the value reported by Lurie (17). At 5740 feet, he found an average value of 5.593 million cells per cubic millimeter in the blood of young adult males. His subjects were younger and the altitude slightly higher than in this study. The average red cell count and hematocrit found in the present study were higher than those reported in other studies of young adults which have been reviewed. Part of the difference can be accounted for by reason of the fact that most of these studies were conducted with women who normally have lower values. But the difference cannot be accounted for entirely in this manner. The difference in altitude probably is the one most influential factor.

The results of this study do not agree with the results of other studies among the aged. Using Osgood's and Wintrobe's normal standards of 4.6 to 6.2 million cells per cubic millimeter (average 5.4 million),

Miller (19) found that values for his aged subjects were slightly low, (range 3.5 million to 5.5. million cells, with an average of 4.6 million). All of his subjects were males. The values that Stieglitz (39:818) gave are almost identical with those of Miller (19). In a study by Fowler et al. (11), the average erythrocyte count for males (4.61 million cells) was comparable to the values of Miller (19). The mean hematocrit was stated as 41.7 percent of the total blood volume. Rourke et al. (35) stated the normal hematocrit range as 40-54 percent. According to Phillips (25:Figure 3) normal hematocrit values for men range from 42 to 51.5 percent. This classifies Fowler's value as low, but within normal limits. Newman and Gitlow (23) found the mean values among aged men for erythrocyte count was 4.42 million and for hematocrit 41.2 percent. Their erythrocyte value is lower than those in the findings above among aged subjects, but the hematocrit value is comparable to that found by Fowler et al. (11). In the present study, the mean values for erythrocyte count (5.556 million) and hematocrit 49.0 percent were considerably higher than those values in investigations of the aged quoted above. These differences between the values found in this study and other studies of the aged may be because of the difference in altitude and probably partially because of the health of the individuals. It would seem that active,

independent men might have higher blood values than men in institutions and hospitals by virtue of the fact that they are healthy enough to live by themselves.

It was found that there was considerable color fading within the first hour when determing hemoglobin by the described method. The method (Storvick, 40) stated that accurate results could be obtained within the first three hours. However, toward the end of the experiment, it was noticed that values were higher if read sooner. Further study showed that hemoglobin samples did not fade consistently at the same or different concentrations. Therefore, no accurate correction could be made for fading. The last few samples were read immediately. Subjects could not be brought to the laboratory again for re-determination of the hemoglobin because of the difficulty in contacting them. The mean of the hemoglobin values as determined uncorrected for the study was within normal limits but toward the lower end of the range. Correction to approximate the true values would raise the average considerably. No individual values at either sampling were markedly low. The values for the last few samples which were read immediately were consistently high. From these findings, it is probable that the hemoglobin values for these aged subjects are within the normal range.

The specific gravity of the plasma in the

present study was found to be markedly low according to Phillips (25:Figure 3). His normal range was set from 1.0255 to 1.0283. The mean value for this study was 1.0212. Since none of the subjects suffered from any serious debilitating disease to the best of their knowledge, these diseases would probably not be the primary factor accounting for the low values. None of the subject's "typical" diets showed a marked lack of protein. However, most of the diets were classified as Grade 3 or poor diets and the possible effect on the plasma protein is not known. The few subjects that had good diets did not have higher plasma protein values. Hypoproteinemia is common in serious kidney disease. Stieglitz (39:659) stated that in aged subjects hypoproteinemia is most frequently caused by digestive disorders, anorexia, nausea and vomiting, and injudicious limitation of dietary protein. He spoke of these causes as contributing to edema in acute nephritis. Whether or not such minor non-chronic illness alone can cause hypoproteinemia in older people in supposedly good health is questionable. Since no test for proteinuria was made in this study, its influence is unknown. Hypoproteinemia may be an accompaniment of the aging process and influenced by poor absorption of food. Since studies of hypoproteinemia in aged subjects have not been discovered in the literature by the writer, it seems that further

study in this field is necessary.

Todd and Sanford (41:439) considered 0.9 to 1.5 milligrams of ascorbic acid per 100 milliliters of plasma a normal value. Donelson et al. (8), Purinton and Schuck (27), Fincke and Landquist (10), Prunty and Vass (26) used 0.8 milligrams percent as normal. McMillan and Todhunter accepted 0.80 milligrams or above as an adequate state of ascorbic acid nutrition and below 0.40 to 0.50 milligrams percent as a state of moderate depletion of tissue stores and below 0.4 milligrams a state of fairly marked depletion but not tissue exhaustion. Rourke, et al. (35) considered 0.4 to 1.0 milligrams percent for fasting blood as "normal". This may be "normal" in that the majority of values fall within this range but in the lower limits can hardly be indicative of tissue saturation. Accepting 0.8 milligrams and above as normal, 0.4 to 0.8 as a depletion, and below 0.40 as marked depletion, the mean value, 0.61 milligrams percent, for ascorbic acid in the plasma in this study is low. However, it is comparable to the mean value of 0.66 milligrams percent of young adults found by Donelson et al. (8), the mean of 0.67 milligrams percent for college freshmen found by Dodds and MacLeod (5), and 0.65 milligrams by McMillan and Todhunter (18). The elderly subjects of Rafsky and Newman (33) had been on a good diet in an institution for some time. This more or

less controls dietary habits and accounts in part for the higher mean value of 0.72 milligrams percent before the saturation tests began. Still the findings of all these studies reveal values below the accepted standard for adequate ascorbic acid nutrition. The study of Rafsky and Newman (33) may indicate that aged people on a good diet have lower plasma ascorbic acid values than expected by reason of the lowered absorption and utilization and the general slowing down of the bodily processes.

This study was conducted in the early spring in a season in which there were few fresh vegetables and fruits on the market and most of them were quite expensive. This unavailability of cheaper fruits and vegetables rich in ascorbic acid accounts in part for the low values of the subjects. Over half of the subjects had no citrus fruit or tomatoes in their "typical" diets and tended to have lower plasma ascorbic acid values than the other subjects. These poor diets may be, in part, explained by the unavailability of fresh fruits and vegetables and partly because of poor dietary habits. Although extreme care to prevent hemolysis was taken, it sometimes occurred and might account for the lowering of the values (Simmons and Gentzkow, 37:248).

It was found that the mean erythrocyte counts and hematocrit values of this study were within the

upper margins of the normal range for adults at lower altitudes. The mean plasma specific gravities and reduced ascorbic acid values were markedly below the normal range for young adults.

From this study with aged subjects it seems improbable that a mass study of aged subjects could be conducted on "assembly-line" basis as easily as similar studies with young adults. Each subject requires a good deal of time, especially for interviewing. Old people, living alone as these subjects were, had an abundance of time and could not be hurried without sacrificing their cooperation. Therefore, this type of subject must be handled with extreme care if his cooperation is to be gained and maintained.

The actual blood taking could be done rapidly. Methods used in analyzing the blood were found to be feasible and satisfactory for this age group. However, further work is needed in standardizing the hemoglobin method and in comparing the micro-method with the macro-method for hematocrit.

This study was intended to be of a preliminary nature and further study on a great number of subjects is needed in order to establish normal values for blood constituents in elderly people of known dietary history living at higher altitudes.

Chapter VI

SUMMARY

Since there is a disproportionate increase in the number of older people in this country and few studies on their nutritional status have been carried out, this study was planned. It also seemed of importance to test some newer methods of nutritional investigation on this age group.

The object of this experiment was to determine the values for selected factors of nutritional significance in the blood of men aged 70 to 80 living at 5000 feet altitude. Health histories and diet records were also to be obtained to help in the interpretation of the data.

Two samples of blood from each of 25 subjects were analyzed for hemoglobin, erythrocyte count, hematocrit, plasma ascorbic acid and plasma specific gravity by methods applied to fingertip blood. The samples were taken a week apart. The mean values were: red cell count 5.556 million cells per cubic millimeter of blood; hematocrit, 49.0 percent; ascorbic acid, 0.61 milligrams per 100 milliliters of plasma, and plasma specific gravity, 1.0212. The mean values for erythro-

cyte count and hematocrit, were higher than those for young or aged subjects at lower altitudes. Hence, altitude is probably one of the factors which explain the higher values obtained.

The mean values for ascorbic acid of the plasma were comparable to, although somewhat lower, than the values for young adults as found in other surveys. However, the mean ascorbic acid value was lower than that of aged subjects on a controlled good institutional diet. Poor food habits are partially responsible for the low plasma ascorbic acid values of the studies subjects, especially since many of the subjects did not eat citrus fruit or tomatoes. The mean value for the specific gravity of the plasma was below the normal range for young adults. These values could not be explained by the incidence of serious chronic diseases.

Mass nutritional study of aged subjects needs more careful planning and more time than similar studies among younger subjects, but are still feasible.

Further study of the blood constituents of aged subjects living at 5000 feet altitude is indicated by this study.

A P P E N D I X

TABLE OF CONTENTS

Appendix A.--FOOD RECORD SHEET

Name _____ Date _____
Month Day Year

Adress _____ Age _____ Sex _____
Years Months

FOODS EATEN ON _____
Day of week

Food	Amount	Food	Amount
BREAKFAST			
BETWEEN BREAKFAST AND NOON MEAL			
NOON MEAL			
BETWEEN NOON AND EVENING MEAL			
EVENING MEAL			
AFTER EVENING MEAL			
SUPPLEMENTS			

Appendix B.--SUMMARY OF MENU

BASIS: Basic 7 (Recorded in servings; ½ cup taken as serving for cooked fruits and vegetables.)

Whole grain or enriched cereal products _____
Potatoes _____
Green or yellow vegetables _____
Other vegetables _____
Citrus fruit or tomatoes _____
Other fruit _____
Meat or meat substitutes _____
Milk _____
Eggs/ week _____
Fat (enriched) teaspoons of _____

GENERAL QUESTIONS:

Number of meals/ day _____
Glasses of water/ day _____
Meal intervals_____Regularity of meals_____
Food dislikes, allergies, idiosyncracies

Eating difficulties

Vitamins

Laxatives

Medicines

Appendix C.

NO.
Date

NUTRITION STATUS STUDY
COLORADO
HEALTH RECORD

Name
Age Place of birth Height Weight

Condition of:

eyes

teeth

skin

hearing

Degree of activity

Sleep-av. no. hrs./day sound or fitful

Exercise - kind amount

Symptoms of malnutrition:

Incidence of disease

	Present	
Severity	Duration	Comments

	Past	
Severity	Duration	Comments

Appendix D. HEALTH RECORD GUIDE SHEET
(Not to be written on)

Condition of:

eyes
- abnormal discharge of tears
- night blindness
- abnormal intolerance of light
- burning or itching of eyes
- spots in front of eyes

teeth
- sore bleeding gums
- cavities
- dentures
- no. of teeth condition of:

skin
- dry and wrinkled
- color
- burning or prickling of skin

hearing
- ability to hear

Symptoms of malnutrition:
- lack of appetite
- lassitude or chronic fatigue
- recent loss of weight
- lack of mental application
- loss of strength
- nervousness and irritability
- burning or prickling of skin
- muscle and joint pains

Incidence of disease
- Hay fever
- Pneumonia
- Colds
- Sinusitis
- Influenza
- Abnormal blood pressure
- Diarrhea
- Constipation
- Frequent digestive disturbances
- Rheumatism or arthritis or others
- Nephritis
- Hepatitis
- Tuberculosis
- Heart disease
- Diabetes
- Peptic ulcer Cancer

Appendix E.--INDIVIDUAL VALUES FOR THE BLOOD CONSTITUENTS OF ALL SUBJECTS CLASSIFIED BY AGE

Subject number	Age years	Erythrocytes million		Plasma (1)		Hematocrit percent		Ascorbic acid (2)		Menu evaluation
Samplings		1st	2nd	1st	2nd	1st	2nd	1st	2nd	
2	70	4.94	5.29	--	1.020	51	49	--	1.22	3
17	70	5.47	5.91	--	1.020	49	--	0.14	0.18	3 #
9	70	6.28	4.76	1.023	1.020	48	50	0.96	0.52	3 #
25	71	4.45	5.53	--	1.020	45	--	--	0.18°°	3
30	71	6.19	5.39	1.024	1.020	50	49	--	1.19	3 #
24	72	4.89	5.89	--	1.023	52	42	0.84	--	1
27	72	5.25	6.59	--	1.017	57	54	--	0.43°	3 #
23	72	5.34	5.66	--	1.019	50	--	0.20°	1.80	3 #
22	72	6.17	--	1.020	--	48	--	0.58°	--	3 #
16	72	5.89	4.63	1.020	1.021	48	50	0.10°	0.40	2 #
32	72	6.22	6.61	1.024	1.020	56	--	1.01	1.22	2
20	73	5.38	5.78	--	1.023	51	45	--	--	3 ##and#
13	73	5.18	5.28	1.020	1.018	49	44	1.45°	1.26	3
14	73	6.20	6.43	1.020	1.020	47	42	0.86	0.78	3
29	74	4.60	5.83	--	1.020	45	42	0.87	1.01	3
4	74	6.82	5.53	--	1.023	47	47	--	0.47	3 #
1	74	6.09	5.49	1.024	1.023	48	47	0.61°	0.94	3 ##
21	76	4.48	5.76	--	1.017	44	52	o.11	0.29	3 #
7	76	4.87	3.73	1.020	1.023	49	49	--	0.52	3 #
31	76	5.82	5.24	1.024	1.024	46	46	0.96	0.24	1
12	76	6.15	5.05	1.020	1.020	50	45	0.29	0.27	3 #
3	77	5.19	6.80	--	--	59	--	--	0.43	3
28	77	4.64	5.92	1.024	1.023	48	50	0.58	0.22°	3 #
5	78	4.94	5.33	--	1.024	--	53	--	0.11°	3
6	78	5.46	5.82	--	1.028	48	45	0.04°	0.25°	3 #

\# No citrus fruit or tomatoes
Restricted diet.
° Slight hemolysis (1) Specific gravity
°° Hemolysis (2) Milligrams per 100 milliliters of plasma.

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