Relationships Between Coronary Heart Disease Risk Factors And Serum Ionized Calcium In A Kennedy Space Center Cohort

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Relationships Between Coronary Heart Disease Risk Factors And Serum Ionized Calcium In A Kennedy Space Center Cohort

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ABSTRACT

INTRODUCTION: Kennedy Space Center (KSC) employees are reported
to be at high risk for coronary heart disease (CHD). Risk factors
for CHD include high serum total cholesterol levels, low levels of
high-density lipoprotein cholesterol (HDLC), elevated triglyceride
values, cigarette smoking, physical inactivity, high blood
pressure, being male, and being older. Higher dietary and/or
serum calcium may be related to a lower risk for CHD. We
evaluated these risk factors for CHD in a cohort of KSC employees
and examined their relationships with serum ionized calcium (Ca^{++})
levels. METHODS. Fifty men and 37 women participated. Subjects
were tested in the morning after fasting 12 hours. Information
relative to smoking and exercise habits was obtained; seated blood
pressures were measured; and 24 ml of blood were drawn. RESULTS.
KSC men had higher risk values than KSC women as related to HDLC
(men 39.2 mg/dl; women 60.1 mg/dl), triglycerides (men 121.0
mg/dl; women 80.2 mg/dl), systolic blood pressure (men 119 mmHg;
women 109 mmHg), and diastolic blood pressure (men 81.5 mmHg;
women, 74.2 mmHg). Smoking and non-smoking groups did not differ
for other risk factors or for serum Ca++ levels. Exercise and
sedentary groups differed in total cholesterol and triglyceride
levels. Serum Ca^{++} levels were related to age, and this Ca^{++}/age
relationship differed between regularly exercising subjects and
sedentary subjects. In the sedentary group, serum Ca^{++} levels
increased with increasing age; in the exercisers, serum Ca^{++}
decreased with increasing age. These relationships did not differ
between the men and the women. CONCLUSION. These relationships
may be significant to the risk of CHD and/or the risk of bone
demineralization in an aging population. Further research is
indicated.

INTRODUCTION

Kennedy Space Center (KSC) employees have been reported to be at a greater risk of developing heart disease than the average population ([23]). Thus, information which aids in understanding this risk is of potential importance to NASA. Among the reported factors predictive of coronary heart disease (CHD) risk in the general population are high serum cholesterol levels, low levels of high-density lipoprotein cholesterol (HDLC), elevated triglyceride values, cigarette smoking, physical inactivity, and high blood pressure ([21]). Men have a greater risk of CHD than women, and risk appears to increase with age ([1,8,11]). Recent reports suggest a role for calcium as a protective factor in modulating risk of CHD ([1,3,18,19]).

Cholesterol, triglycerides, and phospholipids are classified together as lipids. They are transported in the blood by chylomicrons, very-low-density lipoproteins (VLDL), low-density lipoproteins (LDL), high-density lipoproteins (HDL), and serum albumin. In persons without lipoprotein abnormalities, approximately 70 percent of plasma cholesterol is transported by LDL; thus the total plasma cholesterol level largely reflects the LDL cholesterol levels. Total cholesterol and LDL cholesterol are related to development of CHD. Elevated levels of plasma HDLC, on the other hand, are associated with a lowered risk for developing CHD ([13]). Data from one study provided evidence of a strong negative association of HDLC level with the subsequent incidence of CHD in both men and women over age 50 ([12]). The American Heart Association reported that the magnitude of the negative association of HDLC with CHD within a wide range of geographic, ethnic, social, age, gender, and race groups has been found to be as large as or larger than that of the other known risk factors ([26]). High-density lipoprotein is thought to be a "cholesterol scavenger" which interferes with the atherosclerotic process by removing cholesterol from the plaque inside the arterial wall and transporting it to the liver, where cholesterol is converted to bile acids and excreted via the intestine ([11]).

Cigarette smoking is a major risk factor for CHD; however, the mechanisms of the deleterious effects are still unclear. The increased carbon monoxide in blood may damage the endothelium and accelerate the entry of cholesterol into the wall of the arteries. Nicotine may induce cardiac arrhythmias. Smoking also enhances platelet aggregation ([4]). In one study of cigarette smokers from randomly selected families, the smokers had significantly lower HDLC levels and higher VLDL and plasma triglyceride levels than ex- and non-smokers ([4]).

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A study of 15,936 Harvard male alumni revealed that lower levels of daily energy expenditure were associated with an increased risk of cardiac death ([22]). One mechanism of decreased risk in physically active individuals may be activity-induced changes in lipids. The possibility of producing a favorable alteration in blood lipid profiles through formal exercise programs has been investigated in several studies. In one study exercise lowered total triglyceride and total cholesterol levels, increased HDLC, and improved cholesterol/HDLC ratio ([21]). In another study, three exercise groups were compared with regard to HDLC levels. The groups were marathon runners, joggers, and inactive men. The researchers suggested that differences in HDLC levels were related to exercise habits instead of diet ([13]). Many studies have verified the advantageous effect of increased physical activity on plasma lipoproteins in men ([27]). However, alterations occur only after a considerable amount of regular, vigorous exercise ([16]). Furthermore, exercise conditioning leading to improved physical fitness in healthy women may not be associated with increments in HDLC levels. In two ten-week studies of healthy, sedentary women, values of HDLC did not differ after a rigorous exercise program ([9],[28]).

Hypertension is one of the major risk factors of CHD. The cause of hypertension is often not known, but it may be associated with renal pathology, atherosclerosis, Na+ intake, or psychological stress. Hypertension places a strain on the heart by requiring it to pump blood against an increased pressure; this could lead to heart failure. High blood pressure may also cause rupture of small blood vessels.

Blood calcium levels may be related to these risk factors. Fifty percent of total calcium in serum is ionized while the remaining calcium is protein bound. The physiological importance of extracellular ionized calcium on cardiac function was first established by McLean (1935), who observed that the amplitude of contraction of the isolated frog heart was proportional to the ionized fraction of calcium in serum ([15]). Calcium ions are also essential for the preservation of skeletal structure, muscle function, activation of several enzymes, blood coagulation, and transmission of nerve impulses ([14]).

Several studies have shown that dietary calcium is inversely related to blood pressure. In one study, 57 subjects who received one-gram supplements of elemental calcium daily for 22 weeks experienced a significant decrease in diastolic blood pressure ([3]). In another study hypertensive individuals were found to consume significantly less calcium in their diets than their normotensive counterparts ([20]).

The purpose of this study was to evaluate relationships
between serum ionized calcium levels and other risk factors
for CHD in a subset of KSC employees.

METHODS

Eighty-seven employees of the KSC, 50 men and 37 women aged
43.6 ± 11.3 (S.D.) years, participated in this study. Each
subject visited the Biomedical Laboratory in the morning.
Subjects were asked to refrain from eating and drinking
(including alcohol) twelve hours prior to their visit.

Qualified laboratory personnel interviewed the subjects,
and the answers to certain pertinent questions helped to
classify the subjects into appropriate exercise and smoking
groups. According to the physical activity that they
performed on a regular basis, the subjects were placed in
the active, the moderately active, or the sedentary group.
Active subjects participated in regular programmed
activities; they performed strenuous aerobic exercise at
least three times per week for 30-minute intervals.
Moderately active subjects participated in occasional
physical activities which were generally not considered
aerobic. The type of exercise for the active and
moderately active subjects was recorded. Sedentary
subjects performed no exercise.

The subjects were also classified into three groups
according to cigarette smoking: subjects who smoked
regularly, subjects who had never smoked, and subjects who
had stopped smoking. Subjects who had stopped smoking
three months or more prior to the time they visited the
laboratory were considered in the latter group; subjects
who stopped smoking less than three months prior to the
visit were considered in the smoking group.

After the subjects sat quietly with uncrossed legs for at
least five minutes, with the right arm at the level of the
right atrium, blood pressure was monitored using a mercury
sphygmomanometer. Heart rate was then monitored and
recorded, after which a second blood pressure reading was
obtained.

Twenty-four ml of blood were obtained from an antecubital
vein. Within one hour of collection, heparinized whole
blood was analyzed on a Nova II Ion-Selective Electrode
Analyzer for ionized calcium. The Nova II utilizes direct
potentiometry, an analytical method for measuring
electrolytes in undiluted samples [25]. Hemoglobin was
determined using a Coulter Hemoglobinometer after diluting
a portion of anticoagulated blood with disodium

ethylenediaminetetraacetate (EDTA) and treating it with Zapoglobin II, a lysing agent ([6]). The hemoglobinometer is a photo-electric system coupled with integrated circuits ([5]). A portion of blood was placed in two capillary tubes and spun in a centrifuge. Hematocrit was determined using a micro-capillary reader. The DuPont ACA III discrete chemistry analyzer, which uses a photometer process was used for enzymatic analyses of triglycerides, cholesterol, and HDLC ([7]).

Data were entered into an IBM PCXT computer and analyzed using Systat ([24]). The following statistical tests were performed:

1. Means and standard deviations of all continuous variables for the total sample and all classification subgroups;

2. T-tests of all variables comparing male and female subgroups;

3. Correlational analyses among all continuous variables for the total sample and for male and female subgroups;

4. One-way analyses of variance (ANOVA's) for all continuous variables contrasting exercise groups, alcohol groups, and smoking groups;

5. Test of differences among exercise group correlations of calcium ions and age;

6. Test of differences in age and calcium ion correlations between genders;

7. Correlations between ionized calcium and age for males and females separately.

RESULTS

Means and standard deviations of all continuous variables are reported for the total sample (Table 1) and for the male and female subgroups (Table 2). The men had lower HDLC values, higher triglyceride values, higher blood pressure values and higher hemoglobin and hematocrit values than the women. Mean age of male and female subgroups were made commensurate with each other by subject selection. The nonsignificant age difference for genders was a confirmation of this strategy.

Significant correlations among continuous variables are listed in Table 3.

The population breakdown for exercise and cigarette smoking is presented in Table 4.

Significant ANOVAs were obtained for total cholesterol and triglycerides when contrasting exercise groups (Table 5); no significant ANOVAs were obtained when contrasting cigarette smoking groups.

The regression of calcium ions on age was significant for the sedentary and active groups, but not for the moderately active group. For sedentary subjects, an increase in age of one year was on the average associated with an increase of 0.017 mg/dl of ionized calcium (Figure 1); for moderately active subjects a one-year increase in age was associated with a 0.004 mg/dl decrease in ionized calcium (Figure 2); while for active subjects a one-year increase was associated with a 0.006 mg/dl decrease in ionized calcium (Figure 3). Significant differences between the correlations of age and ionized calcium exist between the moderately active group and the sedentary group ($z = -2.856$) and between the active group and the sedentary group ($z = -4.263$). There were no differences in correlations of these variables between the male and female subgroups; and separate correlations between ionized calcium and age for men and women were not significant (men, $p=0.18$; women, $p=0.39$).

DISCUSSION

In this study, we have confirmed in a group of KSC employees some well-known relationships concerning CHD risk factors. These data have special significance, since it has been reported that KSC employees are at a greater risk of coronary heart disease than the average population in the United States [23]. We have also revealed some new relationships concerning serum ionized calcium and physical activity level.

Men have been reported to be at a higher risk for CHD than women [2,8,11], and this relationship appears to hold for our subject population. High density lipoprotein cholesterol is inversely related to CHD risk [12], and in this group of KSC employees the men had significantly lower levels of HDLC than the women. The men also had higher values of triglycerides and higher blood pressure values than the women. Our results are also in agreement with reports that men have higher hemoglobin and hematocrit values [15].

Of the correlations among variables in this study (Table 3), several were predicted, and our data confirm other studies: cholesterol with triglycerides, HDLC with

triglycerides, systolic blood pressure with diastolic blood pressure, and hemoglobin with hematocrit. Other significant correlations among variables were not expected and may provide the basis for future research: hemoglobin with HDLC, hemoglobin with triglycerides, hematocrit with total calcium, hematocrit with HDLC, and hematocrit with triglycerides.

Several studies have indicated that dietary calcium intake is inversely related to blood pressure ([17],[18],[19],[20]). We investigated the relationship between blood calcium levels and blood pressure by correlational analysis and found that blood pressure was not significantly related to either total calcium or ionized calcium in our subject population.

The failure to find significant correlations between cigarette smoking and the other risk factors studied in this population may be due to the relatively small size of our sample.

With regard to activity level, our findings that cholesterol and triglycerides differed significantly among groups is in agreement with the published literature ([27]). However, the exercise-related elevations in HDLC which have been suggested by other studies ([8],[13]) were not shown in this study (Table 5).

Our major new finding relates to the relationship of serum ionized calcium with age and exercise levels. Serum ionized calcium was not related to exercise level as examined by a one-way analysis of variance. However, the relationship between ionized calcium and age was significantly different across the exercise categories. Thus, the level of ionized calcium within an exercise category appears to depend upon the age of the subject. The increase in serum ionized calcium with age in the sedentary group could result from increased bone demineralization with older age. This effect was not seen in the groups which performed some exercise and may have been reversed. Because of the age-related bone loss in women due to osteoporosis ([10]), we tested our data for gender differences in the relationship between age and calcium ion, but no differences were revealed.

Further investigation must be undertaken to fully understand the implications of the data and to obtain health benefits from them.

In summary, we have identified a relationship between exercise level, blood ionized calcium level, and age. We do not know whether this relationship is significant to risk of CHD, or whether it may be significant to the relationship between age and bone demineralization.

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TABLE 1

VARIABLES FOR TOTAL COHORT

VARIABLES	UNITS	MEAN	STANDARD DEVIATION
AGE	Year	43.6	11.3
IONIZED CALCIUM	mg/dl	4.7	0.2
CALCIUM	mg/dl	9.1	0.3
CHOLESTEROL	mg/dl	201.0	35.9
HDLC	mg/dl	48.4	17.0
TRIGLYCERIDES	mg/dl	104.0	74.5
HEMOGLOBIN	g/dl	14.3	1.2
HEMATOCRIT	%	41.9	3.0
SYSTOLIC BLOOD PRESSURE	mmHg	115.0	13.0
DIASTOLIC BLOOD PRESSURE	mmHg	78.4	8.3

TABLE 2

VARIABLES BY GENDER

VARIABLES	UNIT	WOMEN		MEN	
		MEAN	STANDARD DEVIATION	MEAN	STANDARD DEVIATION
AGE	Year	41.2	11.3	45.2	11.1
IONIZED CALCIUM	mg/dl	4.8	0.2	4.8	0.2
CALCIUM	mg/dl	9.1	0.3	9.1	0.3
CHOLESTEROL	mg/dl	192.0	30.7	206.0	38.4
HDLC *	mg/dl	60.1	17.0	39.6	10.7
TRIGLYCERIDES *	mg/dl	80.2	69.9	121.0	73.4
HEMOGLOBIN *	g/dl	13.2	0.7	15.1	0.9
HEMATOCRIT *	%	39.4	1.8	43.8	2.3
SYSTOLIC BLOOD PRESSURE *	mmHg	109.0	11.9	119.0	12.3
DIASTOLIC BLOOD PRESSURE *	mmHg	74.2	6.8	81.5	8.1

* WOMEN AND MEN DIFFER $p < 0.05$, t-test

TABLE 3

SIGNIFICANT CORRELATIONS BETWEEN CONTINUOUS VARIABLES
FOR TOTAL COHORT

	r	P
CALCIUM/IONIZED CALCIUM	0.28769	0.00836
AGE/CHOLESTEROL	0.22461	0.04121
TRYIGLYCERIDES/CHOLESTEROL	0.33248	0.00213
TRIGLYCERIDES/HDLC	-0.53608	0.00000
SYSTOLIC BLOOD PRESSURE/AGE	0.35137	0.00113
DIASTOLIC BLOOD PRESSURE/HDLC	-0.32686	0.00256
DIASTOLIC BLOOD PRESSURE/TRIGLYCERIDES	0.27416	0.01214
DIASTOLIC BLOOD PRESSURE/SYSTOLIC BLOOD PRESSURE	0.61997	0.00000
HEMOGLOBIN/HDLC	-0.59094	0.00000
HEMOGLOBIN/TRYGLICERIDES	0.39192	0.00025
HEMOGLOBIN/DIASTOLIC BLOOD PRESSURE	0.32123	0.00307
HEMATOCRIT/CALCIUM	0.27116	0.01316
HEMATOCRIT/HDLC	-0.41668	0.00009
HEMATOCRIT/DIASTOLIC BLOOD PRESSURE	0.29223	0.00735
HEMATOCRIT/HEMOGLOBIN	0.91318	0.00000

TABLE 4

POPULATION BREAKDOWN FOR EXERCISE AND SMOKING GROUPS

PHYSICAL ACTIVITY GROUP

	WOMEN	MEN	TOTAL
ACTIVE	13	18	31
MODERATELY ACTIVE	11	23	34
SEDENTARY	13	9	22

CIGARETTE SMOKING GROUP

	WOMEN	MEN	TOTAL
YES	8	9	17
NO	17	28	45
STOPPED	12	13	25

TABLE 5

CHOLESTEROL, TRIGLYCERIDES, AND HDLC ACROSS EXERCISE GROUPS

EXERCISE GROUP	n	CHOLESTEROL* MEAN	SD	TRIGLYCERIDES* MEAN	SD	HDLC MEAN	SD
ACTIVE	31	189.3	34.8	82.6	47.6	48.9	14.5
MODERATELY ACTIVE	34	203.9	33.5	110.7	72.9	45.3	17.1
SEDENTARY	22	211.5	38.0	124.6	99.7	52.5	20.0

* = SIGNIFICANT DIFFERENCE BETWEEN EXERCISE LEVELS AT ALPHA = 0.05

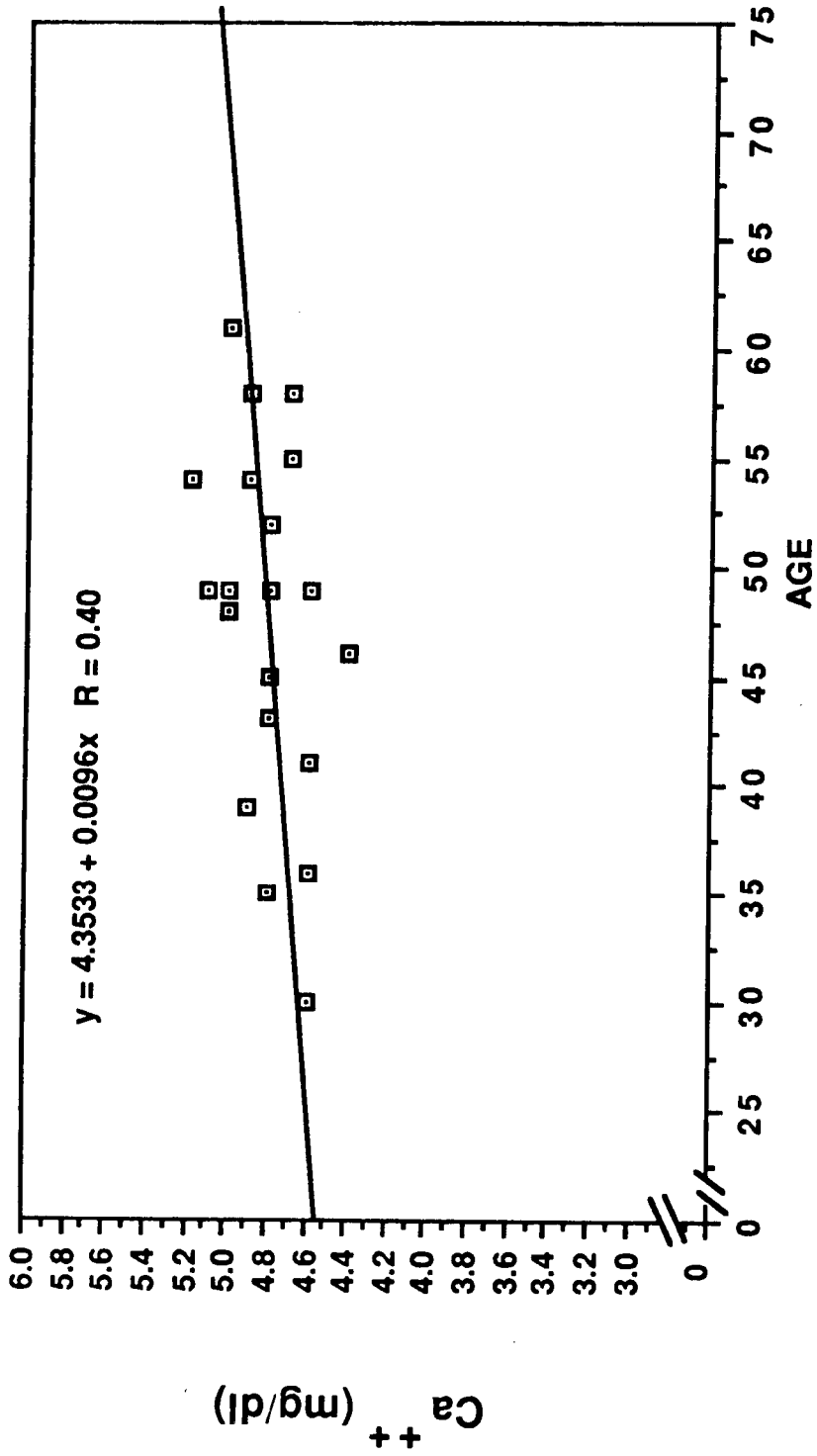SD= STANDARD DEVIATION
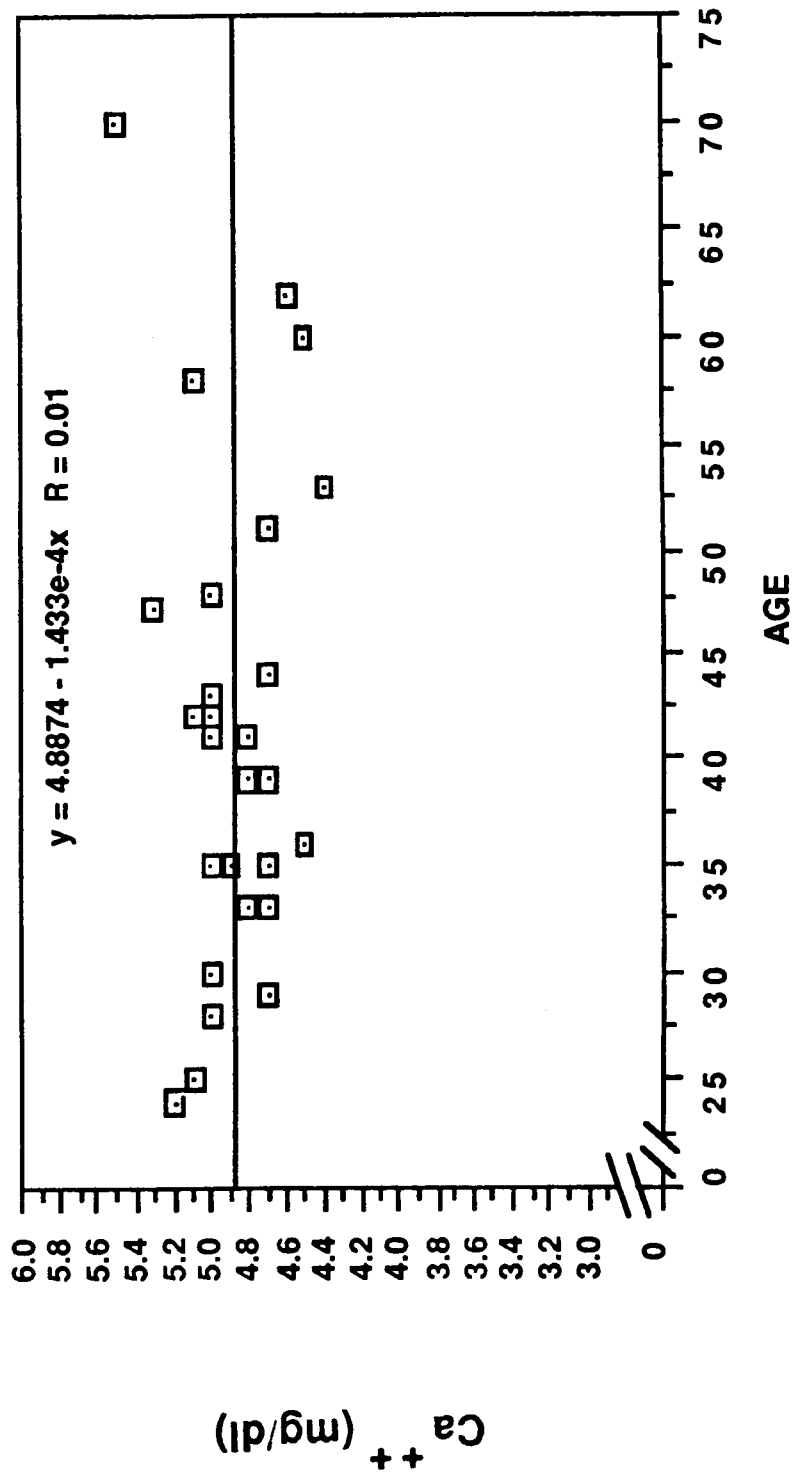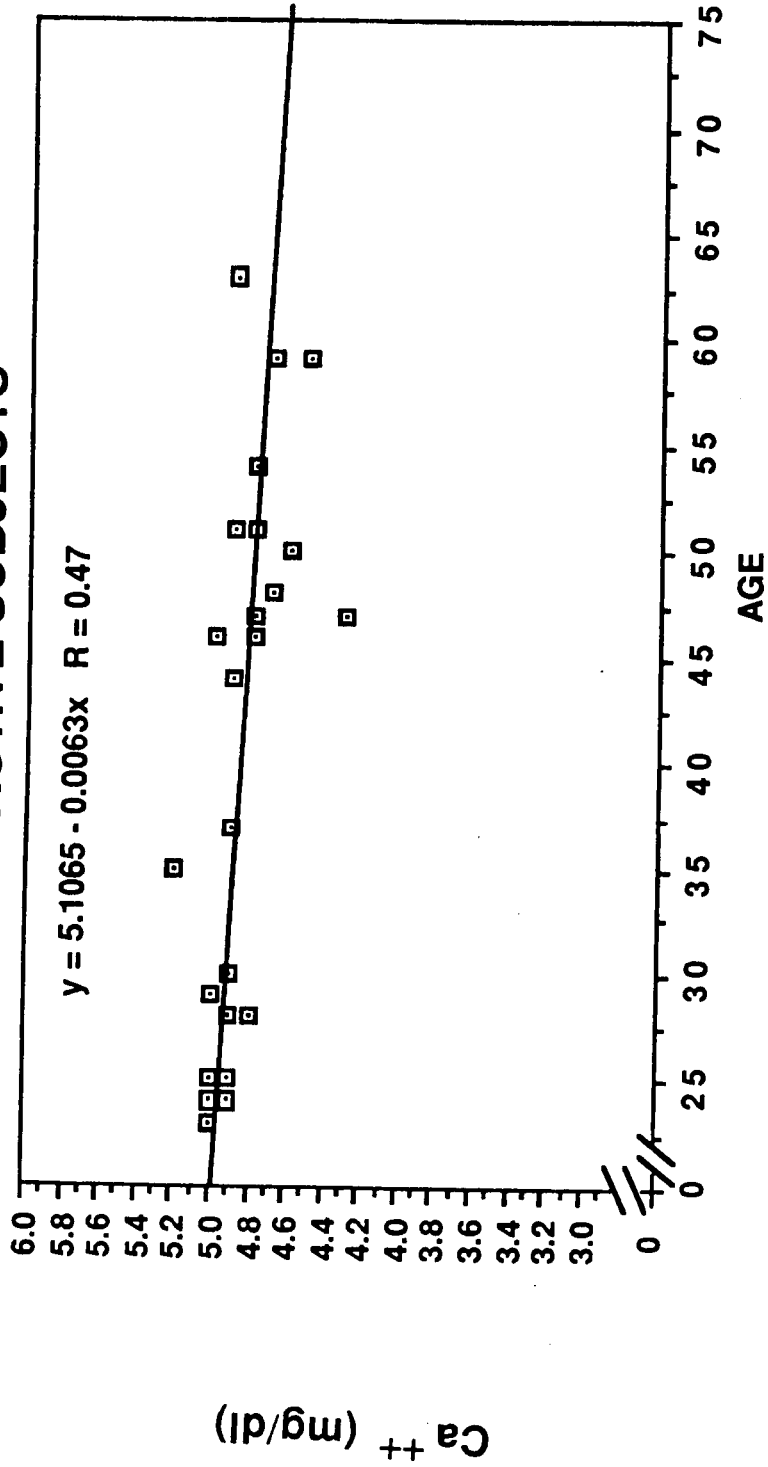
FIGURE 1.
SEDENTARY SUBJECTS

$y = 4.3533 + 0.0096x$ $R = 0.40$

Ca^{++} (mg/dl)

AGE

FIGURE 2.
MODERATELY ACTIVE SUBJECTS

$y = 4.8874 - 1.433e{-}4x$ $R = 0.01$

Ca^{++} (mg/dl)

AGE

FIGURE 3.
ACTIVE SUBJECTS

$y = 5.1065 - 0.0063x$ $R = 0.47$

Ca^{++} (mg/dl)

AGE

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15. Supplementary Notes

16. Abstract

INTRODUCTION: Kennedy Space Center (KSC) employees are reported to be at high risk for coronary heart disease (CHD). Risk factors for CHD include high serum total cholesterol levels, low levels of high-density lipoprotein cholesterol (HDLC), elevated triglyceride values, cigarette smoking, physical inactivity, high blood pressure, being male, and being older. Higher dietary and/or serum calcium may be related to a lower risk for CHD. We evaluated these risk factors for CHD in a cohort of KSC employees and examined their relationships with serum ionized calcium (Ca^{++}) levels. METHODS: Fifty men and 37 women participated. Subjects were tested in the morning after fasting 12 hours. Information relative to smoking and exercise habits was obtained; seated blood pressures were measured; and 24 ml of blood were drawn. RESULTS: KSC men had higher risk values than KSC women as related to HDLC (men 39.2 mg/dl; women 60.1 mg/dl), triglycerides (men 121.0 mg/dl; women 80.2 mg/dl), systolic blood pressure (men 119 mmHg; women 109 mmHg), and diastolic blood pressure (men 81.5 mmHg; women, 74.2 mmHg). Smoking and non-smoking groups did not differ for other risk factors or for serum Ca++ levels. Exercise and sedentary groups differed in total cholesterol and triglyceride levels. Serum Ca^{++} levels were related to age, and this Ca^{++}/age relationship differed between regularly

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16. Abstract (Continued)

exercising subjects and sedentary subjects. In the sedentary group, serum Ca^{++} levels increased with increasing age; in the exercisers, serum Ca^{++} decreased with increasing age. These relationships did not differ between the men and the women.

CONCLUSION: These relationships may be significant to the risk of CHD and/or the risk of bone demineralization in an aging population. Further research is indicated.