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Comparison of antioxidant status  
between young adults and elderly citizens of Giessen,  
Germany

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### O meu contributo para a realização deste trabalho

O trabalho "*Comparison of antioxidant status between young adults and elderly citizens of Giessen, Germany*", foi realizado a partir dos dados dos estudos Giessener Senioren Langzeitstudie (GISELA-study) e Nutrition and health examination study of young adults (NHESYA). Dadas as limitações inerentes ao meu domínio da língua alemã, não me foi possível participar na recolha de dados de projectos de investigação em curso na Faculdade, nem de planear, organizar e realizar um trabalho com população de língua alemã.

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## List of abbreviations

BGA	Bundesgesundheitsamt, Berlin
BMI	Body Mass Index
CHO	Carbohydrate
FDA	Food and Drug Administration
GISELA	Gießener Senioren Langzeitstudie
HDL	High density lipoproteins
LDL	Low density lipoproteins
M	Men
NHESYA	Nutrition and health examination study of young adults
RDA'S	Recommended Dietary Allowances
VERA	Verbundstudie Ernährungserhebung und Risikofaktoren-Analytik (German Nutrition Survey)
VLDL	Very low density lipoproteins
W	Women
Wg	Whole group
WHO	World Health Organization

## 1 Introduction

Since the beginning of 20<sup>th</sup> century the percentage of elderly people has been increasing worldwide, firstly in developed countries, and more recently also in developing ones (WHO, 1998).

To understand the progressive changes in body composition with ageing, the physiological processes behind these changes and also the nutritional needs of the elderly are fundamental steps to promote the quality and duration of life of this growing age group.

Ageing is accompanied by a progressive increase in free radical production or/and a progressive decrease in antioxidant protection (Bunker, 1992). Oxidative processes have been implicated in aging and it has been proposed that antioxidants may have beneficial effects on cognitive functions in the elderly (Carr and Frei, 1999). The free radical theory of ageing suggests that progressive defects in protection against free radical reactions allow tissue damage to occur (Bunker, 1992).

An antioxidant is any substance that, when present at low concentrations compared with those of an oxidizable substrate, significantly delays or prevents oxidation of that substrate (Bunker, 1992). Vitamin E, vitamin C, the vitamin A precursor,  $\beta$ -carotene and selenium, are the four dietary antioxidants recognized and accepted by the US Food and Drug Administration (FDA) (Carr and Frei, 1999).

The purpose of this study was to compare the antioxidant profile in two different age groups. Cross-sectional data from *Giessener Senioren Langzeitstudie* (GISELA study) and from the Nutrition and health examination study of young adults (NHESYA) were compared. The objectives were to know if old and young people differ in their intake of antioxidants, if there are serum alterations due to aging and if plasma antioxidants are influenced by intake.

For these purposes, the intake and plasma concentrations of the 4 above mentioned antioxidants, were analyzed and compared with other studies in the field.



## 2 Subjects and Methods

### 2.1 Giessener Senioren Langzeitstudie (GISELA-study)

#### Study design

The *Giessener Senioren Langzeitstudie* (GISELA) is a longitudinal study in an ageing population of Giessen, Germany, in which since 1994 the nutritional and health status of free-living elderly people is investigated in yearly intervals. Within the scope of this study anthropometrical data, body composition, basal metabolic rate, and various biochemical parameters in blood as well as food intake and the corresponding energy and nutrient intake of the study participants are examined. All measurements take place at the Institute of Nutritional Science in Giessen, Germany between June and November of each year. Subjects come to the Institute between 6:00 and 10:00 AM after an overnight fast. The study protocol was approved by the Ethical Committee of the Faculty of Medicine at the Justus-Liebig University Giessen, Germany, and a written informed consent was obtained from each study participant.

#### Subjects

The subjects were recruited by physicians, notices, senior citizens meetings, advertisements in local newspapers as well as by recruitment of friends through subjects who were already participants. Subjects had to be at least 60 years of age, physically mobile, and available around Giessen for the long term. During the first five years of the GISELA study (1994 - 1998), a total of 320 women and 133 men participated in the investigations. The present study includes the cross-sectional data from 103 women and 57 men who participated in the longitudinal study in 1998. Smokers and users of supplements were excluded. Those who did not record their food intake were excluded too.

## 2.2 Nutrition and health examination study of young adults (NHESYA)

### Study design

Within this study, the nutritional and health status of young adults was investigated for comparison purposes with the elderly population of GISELA. The nutritional and health status of these young people was investigated cross-sectionally in the summer of 1999. Like in the GISELA-study, anthropometrical data, body composition, resting metabolic rate, and various biochemical parameters in blood, as well as food intake and the corresponding energy and nutrient intake of the participants were examined. The measurements took place in the Institute of Nutritional Science in Giessen, during June and July of 1999. The methods that were used within the GISELA-study, were also employed in this study.

### Subjects

The subjects were recruited in May/June of 1999 by advertisements in seminars and papers delivered at the Justus-Liebig-University of Giessen, as well as by recruitment of friends through subjects who were already participants. Subjects had to be between 20 and 35 years-old, without any thyroid dysfunction neither oedema. They shouldn't take any thyroid hormones neither diuretics and the pregnant women were excluded. The sample has 123 subjects (35 men and 88 women), but only 32 women and 8 men were included in the present study, because smokers and users of supplements were excluded, as well as people who did not record their food intake.

## 2.3 Food and nutrient intake

To determine the food and nutrient intake of the subjects a 3-day estimated dietary record was especially developed and validated for the GISELA-study (Lührmann et al. 1999). The dietary record consists of 146 food items, subdivided into 16 food groups. Food items were formed considering the eating habits of the elderly people, which were also assessed by a questionnaire in the GISELA-study. Foods similar in energy and nutrient content were summarized in one food item. These items were analysed by taking the average energy and nutrient content of the single foods. For every food item both typical household measures



(e.g., slice, cup, spoon) and the appropriate weights were given, so with this information the subjects were expected to estimate the amount of their food consumption. The food items were compiled in a record booklet which included instructions with examples. The participants were instructed to record their entire food intake in the diary on three consecutive days, directly after consumption, starting on a Sunday. At the end of the booklet the subjects had the possibility to write down under the heading "others", any consumed food that they were unable to classify.

To analyse the 3-day dietary record first, the foods recorded under the heading "others" were classified among given food items according to their energy and nutrient content. Then data were checked with regard to cooking fat, i.e., if the subjects consumed meat or fish usually prepared with fat, such as cutlet or steak, we checked whether the corresponding fat for frying was also recorded. If reported fat quantities were used as bread spreading only and no cooking fat was written down, fat intake was corrected by adding 4 g cooking oil and 4 g butter for each portion of meat or fish. Then energy and nutrient contents of the food items were calculated by means of the nutrient calculation program CALORA version II and the Federal Nutrient Data Base version II.2 (BGA, 1994).

#### 2.4 Anthropometrical data and body composition

Body weight was measured with a calibrated digital scale (Seca, Vogel & Halke, Hamburg, Germany) to the nearest 0.1 kg after shoes, coats, and sweaters had been removed. For the remaining clothes 0.5 to 1.0 kg were subtracted. Body height was measured in standing position, without shoes to the nearest 0.005 m by a height measurement integrated in the scale.

The Quetelet's Index (QI), also known as Body Mass Index (BMI) was calculated. The index is calculated by dividing the individual's weight (kg) by the square of his or her height (m).

$$\text{BMI} = \text{weight (kg)} / \text{height}^2 (\text{m}^2)$$

Body composition was investigated by using bioelectrical impedance (Akern-RJL BIA 101/S, Data Input, Frankfurt, Germany) in dorsal lying position according to the instruction. Fat-free mass and fat mass were calculated by applying the equation formula from Deurenberg et al (Deurenberg et al, 1991).

## 2.5 Lipid determination in plasma

Blood specimens were collected in heparinized tubes. After drawing and cooling down, the blood samples were centrifuged and the plasma was separated (Heseker et al 1993, Sülteimeier 1996).

The determinations of serum concentrations of total cholesterol, HDL-cholesterol and triglycerides were made with the help of tests of the company „Boehringer Mannheim“, as described below.

### Total Cholesterol

To determine the total cholesterol concentration in serum the CHOD-PAP-Method was employed (Boehringer, 1993). By this enzymatic method, cholesterol esters in a sample are hydrolysed to free cholesterol and fatty acid by cholesterol ester hydrolase. The cholesterol produced together with free cholesterol already present in the sample, is oxidized by cholesterol oxidase (CHOD) yielding hydrogen peroxide. The hydrogen peroxide formed causes phenol and 4 - aminoantipyrine to undergo a quantitative oxidative condensation catalysed by peroxidase (POD), producing a red coloured dye.

The amount of total cholesterol contained in the sample is determined by measuring the absorbance of the red colour.

### HDL-Cholesterol

To analyse the HDL-cholesterol concentration in serum, phosphorwolframic acid and magnesium were used to precipitate the VLDL and LDL-fractions. After a centrifugation, the HDL-cholesterol remains and with the help of CHOD-PAP-Method, as described beyond, is determined (Boehringer 1993).

### Triglyceride

To determine the triglycerid concentration the GPO-PAP method was employed (Boehringer 1993). This is based on the enzymatic hydrolysis of tryglicerides with subsequent determination of liberated glycerol by colorimetry.

### LDL-Cholesterol

The LDL-cholesterol concentration in mg/dl was determined by the formula of Friedewald (1972). It uses the measured tread of total cholesterol (TC), HDL-cholesterol (HDL-C) and



Triglycerid-concentration (TG) like the following formula:

$$\text{LDL-C} = \text{TC} - \text{HDL-C} - \text{TG}/5.$$

### Atherogenic Index

The Atherogenic Index quantifies the proportion of LDL to HDL-cholesterol as a risk factor of Atherosclerosis.

Using the LDL-C calculated by the above formula, and the HDL-C measured concentrations, the Atherogenic Index (AI) was calculated:

$$\text{AI} = \text{LDL-C (mg/dl)} / \text{HDL-C (mg/dl)}$$

### 2.6 Antioxidant determination in plasma

A total of 30 ml blood was drawn by venipuncture for the various blood analysis. For determining plasma concentrations of vitamin E, vitamin C and  $\beta$ -carotene 10 ml blood was collected in EDTA-tubes. For measuring plasma selenium concentrations 10 ml blood was drawn into heparinized tubes. After drawing and cooling down, 4.000 U/min centrifuged the blood samples 10 minutes, and the plasma was separated. Then the samples were stored at  $-80^{\circ}\text{C}$ , prior to laboratory analysis (Gritschneider 1995, Gritschneider et al 1998, Kiel 1997).

Concentrations of vitamin E and vitamin C,  $\beta$ -Carotene and Selenium in plasma samples of subjects were determined in the vitamin laboratory of Institute of Nutrition, Justus-Liebig-University Giessen. Plasma vitamin C concentrations were determined by a modified 2,4-dinitrophenylhydrazine method, which detects ascorbic and dehydroascorbic acid.  $\beta$ -Carotene and  $\alpha$ -tocopherol concentrations in plasma were analysed by a high-performance liquid chromatography method after an extraction procedure with ethanol and hexane.  $\beta$ -Carotene was detected photometrically,  $\alpha$ -tocopherol fluorometrically. Plasma selenium concentrations were determined photometrically. Methods were the same as used in the German Nutrition Survey (VERA-study) and are described in detail elsewhere (Speitling et al 1992).



## 2.7 Statistical analysis

Statistical analysis was conducted using SPSS (Statistical Package of Social Sciences) version 9. Results are presented as median and percentiles 5 and 95. Data analyses included descriptive statistics and U tests.  $P$  values  $< 0.05$  were considered significant. Pearson correlation coefficients were calculated to obtain insight into bivariate associations.

### 3 Results

#### 3.1 Description of the subjects

The sample of this study, is from GISELA study (103 ♀ + 57 ♂), and from Nutrition and health examination study of young adults (32 ♀ + 8 ♂).

##### 3.1.1. Age

Old people are between 60 and 86 years old, and young ones between 20 and 34 years old (Table 1).

**Table 1:** Age in old and young people  
(Median, Percentiles 5-95)

	Old people (n = 160)	Young people (n = 40)
Women (n = 103)	69 62 - 78	24 21 - 33
Men (n = 57)	69 64 - 79	30 22 - 32
Whole group (n = 200)	69 62 - 78	24 22 - 33

##### 3.1.2 Food intake

Food intake data, shows statistically significant differences between age and gender groups in the intake of the various food groups (Table 2).

Young women eat more pasta, rice, muesli and cornflakes than older ones, which is also observed for men. Old men eat more fruit and fruit products than younger ones but young men eat more vegetables than old ones. Older women eat more potatoes, meat and meat product, spread and cooking fat, than younger ones, and young women eat more sugar and sweets and also drink more non-alcoholic drinks than old women.

Fish intake is very low in all groups, with the exception of younger men which showed a median intake of 20g/day.

**Table 2:** Food intake (g/d) in old and young people  
(Median, Percentiles 5 - 95)

		Old people (n = 103 F, 57 M)	Young people (n = 32 F, 8 M)	p <sup>1)</sup>
Bread, buns and cakes	W	180 (70 - 376)	162 (14 - 381)	n.s.
	M	235 (95 - 427)	200 (138 - 258)	n.s.
	Wg	205 (73 - 405)	181 (18 - 376)	< 0.05
Pasta, rice, muesli and cornflakes	W	50.0 (0.0 - 160)	120 (0.0 - 339)	< 0.001
	M	40.0 (0.0 - 202)	163 (0.0 - 400)	< 0.01
	Wg	50.0 (0.0 - 180)	134 (0.0 - 395)	< 0.001
Milk and dairy products	W	203 (52 - 491)	254 (65 - 560)	n.s.
	M	205 (20 - 546)	290 (29 - 878)	n.s.
	Wg	204 (39 - 505)	258 (100 - 560)	< 0.05
Fruit and fruit products	W	238 (72 - 573)	216 (18 - 792)	n.s.
	M	231 (39 - 641)	104 (21 - 405)	< 0.05
	Wg	237 (68 - 583)	178 (21 - 745)	n.s.
Vegetables	W	125 (25 - 393)	184 (16 - 611)	n.s.
	M	113 (23 - 329)	238 (162 - 568)	< 0.001
	Wg	118 (25 - 370)	200 (26 - 566)	< 0.01
Potatoes	W	90 (0.0 - 227)	15 (0.0 - 145)	< 0.001
	M	120 (0.0 - 260)	65 (0.0 - 190)	n.s.
	Wg	100 (0.0 - 239)	20 (0.0 - 187)	< 0.001
Meat and meat products	W	100 (2.3 - 231)	34 (0.0 - 237)	< 0.001
	M	130 (0.0 - 267)	55 (0.0 - 287)	n.s.
	Wg	113 (0.0 - 242)	34 (0.0 - 280)	< 0.001
Fish	W	0.0 (0.0 - 79)	0,0 (0.0 - 56)	< 0.01
	M	0.0 (0.0 - 91)	20,3 (0.0 - 60)	n.s.
	Wg	0.0 (0.0 - 80.0)	0,0 (0.0 - 60)	n.s.
Spread and cooking fat	W	13 (4.0 - 31)	9 (0.6 - 30)	< 0.01
	M	13 (2.0 - 28)	19 (11 - 39)	n.s.
	Wg	13 (4.0 - 30)	11 (1.0 - 34)	n.s.
Sugar and sweets	W	36 (0.0 - 129)	48 (12 - 231)	< 0.05
	M	35 (3.0 - 104)	60 (3.3 - 157)	n.s.
	Wg	35 (0.2 - 123)	48 (8.6 - 204)	< 0.05
Non-alcoholic drinks	W	1250 (500 - 2380)	1968 (833 - 3155)	< 0.001
	M	1250 (395 - 2398)	1508 (450 - 1883)	n.s.
	Wg	1250 (500 - 2350)	1833 (738 - 3071)	< 0.001
Alcoholic drinks	W	42 (0.0 - 327)	0.0 (0.0 - 218)	n.s.
	M	117 (0.0 - 801)	217 (0.0 - 533)	n.s.
	Wg	58 (0.0 - 416)	0.0 (0.0 - 521)	n.s.

W = Women, M = Men, Wg = Whole group

<sup>1)</sup> Mann-Whitney U test



### 3.1.3 Energy, nutrient and cholesterol intake

Although the energy intake was similar across the 4 groups, differences were found in their CHO, protein and fat intakes (Table 3). Younger women have a higher CHO intake than older ones but lower of protein. Comparing the two men groups, differences were only observed in fat intake, which was higher amongst the young ones.

**Table 3:** Energy, nutrient and cholesterol intake in old and young people  
(Median, Percentiles 5 - 95)

		Old people (n = 103 F, 57 M)	Young people (n = 32 F, 8 M)	p <sup>1)</sup>
Energy [kcal/d]	W	1793 (1102 - 3082)	1903 (1166 - 2737)	n.s.
	M	2234 (1191 - 2921)	2344 (2216 - 3503)	n.s.
	Wg	1952 (1111 - 2936)	2008 (1307 - 3254)	n.s.
Carbohydrate [g/d]	W	211 (119 - 342)	245 (139 - 343)	< 0.05
	M	250 (145 - 350)	280 (212 - 435)	n.s.
	Wg	223 (124 - 343)	262 (144 - 364)	< 0.05
Protein [g/d]	W	76 (40 - 124)	63 (28 - 118)	< 0.01
	M	85 (53 - 122)	95 (71 - 162)	n.s.
	Wg	80 (43 - 124)	70 (32 - 143)	< 0.05
Fat [g/d]	W	66 (33 - 128)	63 (33 - 106)	n.s.
	M	85 (35 - 127)	103 (80 - 138)	< 0.05
	Wg	72 (35 - 126)	72 (36 - 124)	n.s.
Carbohydrate [%]	W	49 (35 - 60)	53 (41 - 70)	< 0,001
	M	48 (37 - 58)	45 (37 - 55)	n.s.
	Wg	48 (36 - 59)	51 (39 - 68)	< 0,01
Protein [%]	W	17 (10 - 24)	14 (8 - 24)	< 0,001
	M	16 (12 - 23)	15 (13 - 19)	n.s.
	Wg	17 (11 - 24)	15 (10 - 21)	< 0,001
Fat [%]	W	35 (24 - 48)	33 (21 - 42)	n.s.
	M	34 (25 - 44)	39 (31 - 42)	n.s.
	Wg	35 (25 - 47)	35 (22 - 42)	n.s.

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Cholesterol [mg/d]	W	256 (126 - 510)	227 (34 - 513)	n.s.
	M	313 (158 - 519)	321 (197 - 725)	n.s.
	Wg	272 (129 - 513)	254 (44 - 652)	n.s.

W = Women, M = Men, Wg = Whole group

<sup>1)</sup> Mann-Whitney U test

### 3.1.4 Anthropometric and body composition data

When anthropometric and body composition data in old and young people are compared various statistically significant differences were found (Table 4). Young women have a higher body height than older ones, and this last group have a higher body weight than the young one. The difference of body height between men, old and young, is not statistically significant. However, old men have a higher weight than younger ones.

Old people have higher BMI and fat mass values than young people, in both sexes.

Younger women have higher fat free mass values than older ones. In men no differences were found.

**Table 4:** Anthropometric and body composition data in old and young people

(Median, Percentiles 5 - 95)

		Old people (n = 103 F, 57 M)	Young people (n = 32 F, 8 M)	p <sup>1)</sup>
Body height [cm]	W	161 (149 - 169)	168 (159 - 182)	< 0.001
	M	173 (161 - 184)	178 (168 - 192)	n.s.
	Wg	164 (151 - 181)	170 (160 - 184)	< 0.001
Body weight [kg]	W	69.0 (54.3 - 87.5)	61.5 (48.5 - 69.9)	< 0.001
	M	79.0 (63.9 - 100)	67.8 (59.0 - 81.0)	< 0.05
	Wg	72.0 (56.0 - 95.4)	61.5 (49.5 - 80.2)	< 0.001
BMI [kg/m <sup>2</sup> ]	W	27 (21 - 35)	21 (17 - 25)	< 0.001
	M	26 (23 - 33)	22 (20 - 24)	< 0.001
	Wg	26 (22 - 34)	21 (18 - 25)	< 0.001
Fat free mass [kg]	W	37.3 (29.4 - 45.8)	43.2 (36.1 - 48.1)	< 0.001
	M	52.8 (42.2 - 64.2)	55.0 (50.4 - 65.4)	n.s.

	Wg	41.6 (31.9 - 60.8)	44.3 (36.7 - 64.6)	n.s.
Fat mass [kg]	W	30.5 (22.7 - 44.2)	17.0 (11.4 - 24.2)	< 0.001
	M	25.2 (19.1 - 39.4)	12.5 (7.8 - 16.2)	< 0.001
	Wg	28.8 (20.0 - 43.3)	16.0 (8.7 - 22.7)	< 0.001

W = Women, M = Men, Wg = Whole group

<sup>1)</sup> Mann-Whitney U test

### 3.1.5 Lipid concentration in plasma

Table 5 presents the lipid concentration of the study subjects. Compared to young women, old ones have higher triglycerides concentration in plasma. Total cholesterol is higher in old people than young, for both genders. There is no statistically significant difference in HDL cholesterol concentration in plasma, between old and young people. But LDL cholesterol concentration is higher in old people than young, for both genders. As for total lipids (triglycerides and cholesterol), older subjects of both genders had higher values than younger ones.

**Table 5:** Lipid concentration in plasma in old and young people  
(Median, Percentiles 5 - 95)

		Old people (n = 100 F, 56 M)	Young people (n = 32 F, 8 M)	p <sup>1)</sup>
Triglycerides [mg/dl]	W	108 (57.4 - 271)	78.0 (39.9 - 158)	< 0.001
	M	104 (59.4 - 184)	76.5 (43.0 - 167)	n.s.
	Wg	107 (59.4 - 241)	77.0 (42.1 - 166)	< 0.001
Total Cholesterol [mg/dl]	W	243 (169 - 306)	182 (119 - 276)	< 0.001
	M	214 (121 - 282)	167 (148 - 212)	< 0.01
	Wg	231 (163 - 301)	179 (139 - 266)	< 0.001
HDL Cholesterol [mg/dl]	W	61.7 (35.8 - 83.1)	59.0 (43.3 - 89.3)	n.s.
	M	49.6 (29.1 - 74.4)	46.5 (43.0 - 62.0)	n.s.
	Wg	56.6 (29.4 - 82.0)	58.0 (43.0 - 84.0)	n.s.
LDL	W	152 (95.2 - 218)	102 (42.4 - 190)	< 0.001

Cholesterol [mg/dl]	M	137 (67.4 - 197)	108 (86.0 - 139)	< 0.05
	Wg	145 (90.7 - 216)	104 (55.3 - 175)	< 0.001
Total lipids [mg/dl]	W	353 (266 - 558)	268 (169 - 420)	< 0.001
	M	315 (210 - 452)	247 (201 - 370)	< 0.01
	Wg	341 (247 - 498)	262 (187 - 407)	< 0.001

W = Women, M = Men, Wg = Whole group

<sup>1)</sup> Mann-Whitney U test

### 3.2 Antioxidant status

#### 3.2.1 Vitamin C

Young people of both genders, have a higher vitamin C concentration in plasma than old people (Table 6). In both age groups, women have higher vitamin C concentration than men.

**Table 6:** Vitamin C concentration in plasma (mg/dl) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 156)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 132)	1.27 0.9 - 1.7	1.58 1.0 - 1.8	< 0.001
Men (n = 64)	1.10 0.6 - 1.6	1.33 1.02 - 1.57	< 0.05
p <sup>1)</sup>	< 0.001	< 0.05	
Whole group (n = 196)	1.21 0.7 - 1.67	1.52 1.0 - 1.8	< 0.001

<sup>1)</sup> Mann-Whitney U test

In what concerns vitamin C intake, no statistically significant differences were observed for the two age groups (Table 7).



**Table 7:** Vitamin C intake (mg/d) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 160)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 135)	98 49 - 207	103 46 - 205	n.s.
Men (n = 65)	100 49 - 189	112 54 - 200	n.s.
p <sup>1)</sup>	n.s.	n.s.	
Whole group (n = 200)	100 49 - 206	106 46 - 204	n.s.

<sup>1)</sup> Mann-Whitney U test

No correlation between vitamin C intake and plasma concentration was found in all groups (Table 8).

**Table 8:** Correlation between vitamin C intake (mg/d) and concentration in plasma (mg/dl) in old and young people  
(Spearman)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 200)
Women (n = 135)	R = 0.021 p = n.s.	R = - 0.116 p = n.s.	R = 0.006 p = n.s.
Men (n = 65)	R = 0.121 p = n.s.	R = - 0.333 p = n.s.	R = 0.086 p = n.s.
Whole group (n = 200)	R = 0.061 p = n.s.	R = - 0.153 p = n.s.	R = 0.033 p = n.s.

The correlations between vitamin C concentration in plasma and fat mass (%) is significantly positive for old people and significantly negative for women (Table 9). In the other groups no correlation could be detected.

**Table 9:** Correlation between vitamin C concentration in plasma (mg/dl) and fat mass (%) in old and young people (Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	R = - 0.004 p = n.s.	R = - 0.172 p = n.s.	R = - 0.288 p < 0.01
Men (n = 64)	R = - 0.071 p = n.s.	R = - 0.452 p = n.s.	R = - 0.233 p = n.s.
Whole group (n = 196)	R = 0.241 p < 0.01	R = 0.116 p = n.s.	R = - 0.069 p = n.s.

Between vitamin C plasma and fat mass (kg), there is a significant negative correlation in the whole group and in women (Table 10). No correlations were found in the other groups.

**Table 10:** Correlation between vitamin C concentration in plasma (mg/dl) and fat mass (kg) in old and young people (Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	R = - 0.117 p = n.s.	R = - 0.195 p = n.s.	R = - 0.347 p < 0.001
Men (n = 64)	R = - 0.041 p = n.s.	R = - 0.667 p = n.s.	R = - 0.206 p = n.s.
Whole group (n = 196)	R = 0.050 p = n.s.	R = - 0.034 p = n.s.	R = - 0.215 p < 0.01



### 3.2.2 Vitamin E

As a group, old people have vitamin E concentration values higher than the young ones (Table 11). The differences are statistically significant between old and young, in both genders.

**Table 11:** Vitamin E concentration in plasma (mg/dl) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 155)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 131)	1.38 0.9 - 1.9	1.04 0.8 - 1.4	< 0.001
Men (n = 64)	1.16 0.9 - 1.6	1.00 0.8 - 1.2	< 0.05
p <sup>1)</sup>	< 0.01	n.s.	
Whole group (n = 195)	1.30 0.9 - 1.8	1.04 0.8 - 1.4	< 0.001

<sup>1)</sup> Test Mann-Whitney U

Plasma vitamin E is strongly correlated to blood lipids (Table 7, Table 8, Table 9 and Table 10 appendix). The highest correlation was found between vitamin E and total lipids (Table 12).

**Table 12:** Correlation between vitamin E concentration in plasma (mg/dl) and total lipids concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole (n = 195)
Women (n = 131)	R = 0.575 p < 0.001	R = 0.779 p < 0.001	R = 0.716 p < 0.001
Men (n = 64)	R = 0.662 p < 0.001	R = 0.766 p < 0.05	R = 0.701 p < 0.001
Whole group (n = 195)	R = 0.643 p < 0.001	R = 0.760 p < 0.001	R = 0.728 p < 0.001

Therefore vitamin E was adjusted for total lipids using the residual method (Willet and Stamper 1986, Stryker et al 1988, Heseker et al. 1993) as follows:

$$\text{Vitamin E adj.} = a + b$$

- a: Residual for subject from regression model with vitamin E concentration as the dependent variable and total lipids as the independent variable
- b: expected Vitamin E for a person with mean total lipids concentration.

The adjustment was done on the basis of the following regression line:

$$\text{Vitamin E [mg/dl]} = 0.3817 + 0.0026 \text{ total lipids [mg/dl]}.$$

After this adjustment, statistically significant differences in plasma vitamin E in old and young people were found (Table 13).

**Table 13:** Vitamin E plasma concentration adjusted for total lipids in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 155)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 131)	1.25 0.97 - 1.77	1.20 1.00 - 1.40	n.s.
Men (n = 64)	1.20 0.93 - 1.59	1.26 1.05 - 1.33	n.s.
p <sup>1)</sup>	n.s.	n.s.	
Whole group (n = 195)	1.22 0.94 - 1.74	1.22 1.01 - 1.40	n.s.

<sup>1)</sup> Mann-Whitney U test

Young men have a significantly higher vitamin E intake than old ones (Table 14). Between old and young women no significant differences regarding vitamin E intake were observed. In the younger group, men have higher vitamin E intake than women.

**Table 14:** Vitamin E intake (mg/d) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 160)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 135)	11 6.0 - 21	13 6.4 - 21	n.s.
Men (n = 65)	12 5.7 - 20	17 14 - 23	< 0.01
p <sup>1)</sup>	n.s.	< 0.05	
Whole group (n = 200)	12 5.7 - 21	14 6.5 - 23	n.s.

<sup>1)</sup> Mann-Whitney U test

There is no correlation between vitamin E intake and its plasma concentration in all groups (Table 15).



**Table 15:** Correlation between vitamin E intake (mg/d) and concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 200)
Women (n = 135)	R = - 0.066 p = n.s.	R = 0.245 p = n.s.	R = - 0.035 p = n.s.
Men (n = 65)	R = - 0.074 p = n.s.	R = 0.659 p = n.s.	R = - 0.132 p = n.s.
Whole group (n = 200)	R = - 0.060 p = n.s.	R = 0.248 p = n.s.	R = - 0.062 p = n.s.

### 3.2.3 $\beta$ -Carotene

Table 16 presents the  $\beta$ -carotene concentrations of the study subjects. Compared to old men, young ones have a higher  $\beta$ -carotene concentration in plasma. In women no significant differences were found between the two age groups. Older women had a significantly higher concentration of plasma  $\beta$ -carotene than older men. But no gender differences were observed in the younger group.

**Table 16:**  $\beta$ -Carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) in old and young people (Median, Percentiles 5 - 95)

	Old people (n = 156)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 132)	41 14 - 104	44 20 - 109	n.s.
Men (n = 64)	30 8.0 - 70	52 27 - 152	< 0.05
p <sup>1)</sup>	< 0.01	n.s.	
Whole group (n = 196)	36 12 - 94	46 21 - 127	< 0.05

1) Mann-Whitney U test

In what concerns  $\beta$ -carotene intake younger men had higher intakes than older ones, which were also higher than those of younger women (Table 17).

**Table 17:**  $\beta$ -Carotene intake (mg/d) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 160)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 135)	2.96 0.94 - 9.08	2.89 1.14 - 29.4	n.s.
Men (n = 65)	2.92 0.95 - 12.3	4.67 3.87 - 24.5	< 0.01
p <sup>1)</sup>	n.s.	< 0.05	
Whole group (n = 200)	2.94 0.99 - 9.96	3.57 1.22 - 28.0	n.s.

<sup>1)</sup> Mann-Whitney U test

A positive correlation between  $\beta$ -carotene intake and its plasma concentration was observed for the entire group and for men of both age groups (Table 18).

**Table 18:** Correlation between  $\beta$ -carotene intake (mg/d) and concentration  
in plasma ( $\mu$ g/dl) in old and young people  
(Spearman)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 200)
Women (n = 135)	R = 0.165 p = n.s.	R = 0.133 p = n.s.	R = 0.142 p = n.s.
Men (n = 65)	R = 0.148 p = n.s.	R = 0.571 p = n.s.	R = 0.268 p < 0.05
Whole group (n = 200)	R = 0.160 p < 0.05	R = 0.219 p = n.s.	R = 0.179 p < 0.05

When the correlations between  $\beta$ -carotene and total cholesterol plasma concentrations were analyzed, we found positive significant associations for the entire group of subjects, for the women group, old group and old women (Table 19).

**Table 19:** Correlation between  $\beta$ -carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) and total cholesterol ( $\text{mg}/\text{dl}$ ) in old and young people (Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	R = 0.407 p < 0.001	R = - 0.047 p = n.s.	R = 0.248 p < 0.01
Men (n = 64)	R = 0.137 p = n.s.	R = - 0.024 p = n.s.	R = 0.004 p = n.s.
Whole group (n = 196)	R = 0.389 p < 0.001	R = - 0.054 p = n.s.	R = 0.207 p < 0.01

No correlation was found between plasma  $\beta$ -carotene and total lipids in all groups (Table 17 appendix). Between plasma  $\beta$ -carotene and triglycerides there are significantly negative correlation in the entire group of subject, in old age group, in whole women and old ones (Table 20). In the other groups there are no correlation.

**Table 20:** Correlation between  $\beta$ -carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) and triglycerides ( $\text{mg}/\text{dl}$ ) in old and young people (Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	R = - 0.237 p < 0.05	R = - 0.094 p = n.s.	R = - 0.214 p < 0.05
Men (n = 64)	R = - 0.161 p = n.s.	R = - 0.024 p = n.s.	R = - 0.204 p = n.s.
Whole group (n = 196)	R = - 0.180 p < 0.05	R = - 0.093 p = n.s.	R = - 0.202 p < 0.01



Correlation between plasma  $\beta$ -carotene and BMI is significantly negative in whole group, old age group, women group and old ones (Table 21). In other groups there is no correlation.

**Table 21:** Correlation between  $\beta$ -carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) and BMI in old and young people (Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole (n = 196)
Women (n = 132)	R = - 0.273 p < 0.01	R = - 0.152 p = n.s.	R = - 0.232 p < 0.01
Men (n = 64)	R = - 0.051 p = n.s.	R = - 0.548 p = n.s.	R = - 0.216 p = n.s.
Whole (n = 196)	R = - 0.191 p < 0.05	R = - 0.129 p = n.s.	R = - 0.235 p < 0.01

### 3.2.4 Selenium

Old women have a significantly higher selenium concentration in plasma, than young ones (Table 22). Between men, young and old, there is no difference. In the old age group, women show a higher plasma concentration than men. In the young group no difference between sexes was found.

**Table 22:** Selenium concentration in plasma ( $\mu\text{g/l}$ ) in old and young people  
(Median, Percentiles 5 - 95)

	Old people (n = 155)	Young people (n = 40)	p <sup>1)</sup>
Women (n = 131)	77 51 - 102	66 50 - 96	< 0.001
Men (n = 64)	73 55 - 96	73 56 - 76	n.s.
p <sup>1)</sup>	< 0.05	n.s.	
Whole group (n = 195)	75 53 - 100	68 50 - 96	< 0.001

<sup>1)</sup> Test Mann-Whitney U

Correlation between selenium concentration in plasma and total lipids is significantly positive in whole people and in women (Table 23).

**Table 23:** Correlation between selenium concentration in plasma ( $\mu\text{g/l}$ ) and total lipids concentration in plasma (mg/dl) in old and young people  
(Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = - 0.005 p = n.s.	R = 0.153 p = n.s.	R = 0.188 p < 0.05
Men (n = 64)	R = 0.058 p = n.s.	R = 0.143 p = n.s.	R = 0.087 p = n.s.
Whole group (n = 195)	R = 0.072 p = n.s.	R = 0.155 p = n.s.	R = 0.187 p < 0.01

Correlation between selenium concentration in plasma and fat mass is significantly positive in whole group and in women (Table 24 and Table 25). In the other groups there is no correlation.

**Table 24:** Correlation between selenium concentration in plasma ( $\mu\text{g/l}$ ) and fat mass (%) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 135)	R = - 0.060 p = n.s.	R = - 0.040 p = n.s.	R = 0.208 p < 0.05
Men (n = 65)	R = - 0.147 p = n.s.	R = 0.143 p = n.s.	R = - 0.090 p = n.s.
Whole group (n = 195)	R = 0.089 p = n.s.	R = - 0.146 p = n.s.	R = 0.209 p < 0.01

**Table 25:** Correlation between selenium concentration in plasma ( $\mu\text{g/l}$ ) and fat mass (kg) in old and young people (Spearman)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = - 0.034 p = n.s.	R = - 0.126 p = n.s.	R = 0.218 p < 0.05
Men (n = 64)	R = - 0.068 p = n.s.	R = 0.381 p = n.s.	R = - 0.022 p = n.s.
Whole group (n = 195)	R = 0.045 p = n.s.	R = - 0.149 p = n.s.	R = 0.196 p < 0.01

Correlation between selenium concentration in plasma and protein, expressed as percentage of energy intake per day, is significantly positive in whole group, young people, women, young women and young men (Table 26). In other groups there is no correlation.



**Table 26:** Correlation between selenium concentration in plasma ( $\mu\text{g/l}$ ) and protein intake (% of energy intake/d) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = 0.130 p = n.s.	R = 0.380 p < 0.05	R = 0.269 p < 0.01
Men (n = 64)	R = - 0.055 p = n.s.	R = 0.714 p < 0.05	R = 0.019 p = n.s.
Whole group (n = 195)	R = 0.081 p = n.s.	R = 0.469 p < 0.01	R = 0.198 p < 0.01

Correlation between selenium concentration in plasma and protein expressed as grams intake per day is significantly positive in young people and significantly negative in old people (Table 27). In other groups there is no correlation.

**Table 27:** Correlation between selenium concentration in plasma ( $\mu\text{g/l}$ ) and protein intake (g/d) in old and young people (Spearman)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = - 0.054 p = n.s.	R = 0.289 p = n.s.	R = 0.101 p = n.s.
Men (n = 64)	R = - 0.191 p = n.s.	R = 0.667 p = n.s.	R = - 0.108 p = n.s.
Whole group (n = 195)	R = - 0.161 p < 0.05	R = 0.408 p < 0.01	R = - 0.010 p = n.s.

## 4 Discussion

The purpose of this study was to compare the antioxidant profile in two different age groups. This study shows that there are differences between old and young people in what their intake and/or antioxidant plasma concentration are concerned, and presents some of the reasons for these differences.

It is important to notice that perhaps due to the sample size, most correlations found, although statistically significant, have low values.

### 4.1 Vitamin C

Old people have lower plasma vitamin C values compared to younger ones, but the intakes between both age groups are very similar (Table 6, Table 7). In Germany, in 1987-88 a study on a sample of 2006 healthy people (1144 women, 862 men) aged 18-88 years was carried out. The values found in this study (VERA-study) are considered to be a pattern for comparison of plasma concentrations of antioxidants and vitamins. Our results of vitamin C concentration in plasma, a higher value in young people compared to old, are similar to those found in the VERA-study only in men, as in that study old and young women have a similar concentration (Table 1 and Table 2 appendix). Vitamin C intake, like in our results, is very similar between age groups (Table 3 and Table 4 appendix). The highest concentration of vitamin C was found in young females which is similar to our results.

The lower plasma vitamin C in old people, is in agreement with Bunker (1992), in the work about free radicals, antioxidants and ageing, who explains that elderly may have an impaired absorption and/or poor utilisation of vitamin C. In general, plasma, leucocyte and buffy layer levels of vitamin C in the elderly tend to be lower than in younger ones. Furthermore vitamin C is normally found in high concentration in cells of the immune system and given that there is a decline in immuno competence with increasing age (Hunt et al 1994, Pfitzenmeyer et al 1997), these results were expected.

Blanchard and colleagues studied two groups of healthy women, 14 young aged 20 to 29 years and 14 elderly, aged 65 to 72 years. They also studied two groups of healthy men, 15 young aged 21 to 28 years and 15 elderly aged 66 to 74 years (Blanchard et al, 1990). The purpose of these studies was to compare the pharmacokinetic behavior of vitamin C in a young and an elderly group of healthy, active women and active men, in two states of vitamin



C status, ie, depleted and supplemented. Mean values of BMI were not significantly different in the young and elderly groups. The plasma concentration of vitamin C, when related to body weight, did not differ significantly between states (depleted and supplemented) within age groups since none of the subjects experienced significant weight changes during the study period. Furthermore, the vitamin C concentration in plasma did not differ significantly with age since the mean weights in the two groups were not significantly different. The maximum plasma concentration of vitamin C was not significantly different in the young and elderly groups but was significantly larger in the supplemented state relative to the depleted state in both groups. Cross-correlations were attempted between the young, the elderly, and the combined young and elderly pharmacokinetic parameters and the percentage of body fat and fat-free mass. The data indicate that the rate of absorption is slower in both young and elderly subjects with a higher percentage of body fat. According to Blanchard et al, a possible explanation for this is that vitamin C distributes throughout the body more slowly as the proportion of fat in the body increases. In the present study, correlation between plasma vitamin C concentration and percentage of fat mass, is significantly positive in old people and significantly negative in women. Significantly negative correlations between plasma vitamin C concentration and kg of fat mass in the whole group and in women were also found. Positive correlations found in old people disagree with Blanchard et al studies. However in our results, the elderly have a lower vitamin C concentration and a higher fat mass percentage, compared to the young ones so high fat mass can probably be the explanation for the lower plasma vitamin C in the first group.

There are differences between women and men, in both age groups, despite the same intake, and women have higher vitamin C concentration, like the study by Monget et al (1996). Bunker (1992) considered in his work, and Monget et al (1996) observed in a total of 756 elderly, 193 men and 563 women, that there is a significant gender difference in both plasma and mononuclear leucocyte concentrations of vitamin C. Men have lower levels of vitamin C, but not a lower dietary intake, so a lower renal tubular reabsorption of ascorbic acid in men, can be the reason for the lower plasma vitamin C concentration. Women have higher vitamin C concentration and also higher body fat mass, compared to men.

No correlation between vitamin C concentration in plasma and intake was found. This probably happened because blood samples and food record of the participants were not always obtained at the same time. On the other hand, the vitamin C intake is so high that, perhaps, could not influence concentration in plasma. Some investigators (Nelson et al 1978, Wagner et al 1979) indicated that gastrointestinal absorption pathways of vitamin C become



saturated at doses above 500-1000 mg intake.

Compared to the VERA-study reference for vitamin C concentration in plasma, only a small percentage of our subjects (2.9 % of old women and 7.0 % of old men) were found to have vitamin C concentration below the reference value of 0.65 mg/dl. In the younger group no subjects had vitamin C concentration below the reference value (Table 5 appendix).

In what concerns vitamin C intake below the RDAs reference value of 60 mg/day the highest proportion was found among young men: 25%. In the other 3 groups less than 16% of subjects were found to have a lower ingestion than that (12.6% old women, 12% old men and 15.6% young women) (Table 6 appendix). The mean vitamin C intake is higher for both age groups than the reference value (Table 4 appendix).

#### 4.2 Vitamin E

Old people have a higher vitamin E concentration in plasma but not a higher intake, compared to young people. This age specific difference of vitamin E concentration in plasma is similar to VERA-study results (Table 11 and Table 12 appendix, Table 11), which showed differences between old and young people. In the present study vitamin E intake is higher in young men, which was also found in VERA-study, where old women had a higher intake than young ones and the opposite was found for men (Table 13 and Table 14 appendix). In our subjects young people have a higher vitamin E intake than old people (Table 14).

A positive correlation between vitamin E and age, up to the sixth decade of life, was in agreement with Vandewound et al (1987), in the study about vitamin E status, in which plasma tocopherols and lipids were determined in 95 volunteers. According to Blumberg (1987), serum tocopherol levels increase with age, at least until the seventh decade, in association with the elevation of lipoproteins. Blumberg studied healthy elderly subjects (60-98 years) and a young adult reference group (19-59) recruited from the Greater Metropolitan Boston, Massachusetts area to assess their nutritional status.

Correlation between blood lipids and vitamin E levels has been consistent in many studies. Vandewound et al (1987), concluded that vitamin E plasma levels cannot be interpreted apart from the lipid and especially the cholesterol status. Bunker (1992), when studying free radicals, antioxidants and ageing, considered that plasma levels of vitamin E are strongly correlated with plasma lipid levels, particularly the LDL cholesterol concentration.

In the present study, plasma vitamin E concentration is positively correlated to total

cholesterol, triglycerides and total lipids, the correlation being stronger between vitamin E plasma and total lipids, which include total cholesterol and triglycerides.

Therefore further analysis were carried out adjusting vitamin E for total lipids. No significant differences between the young adults and the elderly subjects were found. These results confirm that the high plasma vitamin E of elderly subjects is due to higher plasma lipids. After this adjustment differences between men and women also lost significance. Elderly women have a significantly higher vitamin E concentration in plasma than elderly men, along with higher plasma lipid levels, with however similar intakes. In the SENECA study, which collected data from 2500 elderly European subjects, speculated that the higher plasma vitamin E concentration in elderly women is probably due to higher cholesterol levels than men and a tendency for these levels to increase in women up to the age of 75-79, whereas it tends to decrease slightly in men after the age of 70 years (SENECA, 1991). The present results also show that the higher vitamin E plasma concentrations are only a result of higher blood lipid concentrations.

There is no correlation between vitamin E concentration in plasma and intake. According to Mensik and Arab (1989), it seems that serum levels of vitamin E are not influenced by its intake. That study analyzed data of the Heidelberg-Michelstadt-Berlin study to see the differences in absolute nutrient intakes, nutrient densities, and nutritional status between more physical active (n = 179) and more inactive people (n = 308).

Younger men in comparison to all other groups, have a higher vitamin E intake. We saw that their intake of spread and cooking fat is higher, however no significant differences between old men and young were found. Furthermore we could find that they have a higher vegetable intake than the other groups, and these intakes can explain the higher vitamin E intake level. Vegetables are the second major dietary source of vitamin E, the first one being fats and oils.

Comparing our subjects vitamin E concentration with the VERA-study reference value, we saw that only a small percentage of them (3.1 % of young women and 1.8 % of old men) have a concentration below the reference value of 0.76 mg/dl (Table 15 appendix).

The mean intake of vitamin E in our subjects is higher than RDAs reference value of 10 mg/day for men and 8 mg/day for women. In old group more subjects (26.3 % of old men and 18.4 % of old women) had a vitamin E intake below RDAs reference value. In young group only a small percentage of women (12.5 %) have a vitamin E intake below the reference value (Table 16 appendix).



### 4.3 $\beta$ -Carotene

Young people have a higher  $\beta$ -carotene concentration, compared to old people. However this higher concentration is a result of the difference found in  $\beta$ -carotene concentration between old and young men. Young men have a higher  $\beta$ -carotene concentration in plasma and a higher intake, compared to old men. Old women have a higher  $\beta$ -carotene concentration compared to old men, despite of a lower intake (Table 16, Table 17).

Compared to VERA-study our young subjects have a higher intake and a higher  $\beta$ -carotene concentration (Table 18, Table 19, Table 20 and Table 21 appendix).

Heseker and Schneider (1994), found plasma  $\beta$ -carotene concentrations strongly associated with BMI. In the present data, there are negative correlations between plasma  $\beta$ -carotene and BMI in the whole group, old people, women and old women. Old women and old men have higher BMI, compared to young women and young men. About  $\beta$ -carotene plasma, old women and men have lower levels, although in women there are not significant differences. With these results we can speculate that old people have a lower  $\beta$ -carotene plasma due to a higher BMI.

Heseker and Schneider in their study, also found that plasma  $\beta$ -carotene concentrations are strongly associated with carotene intake. In our study the correlation between intake and plasma is significantly positive in whole group, old people and in men. Old men have a lower  $\beta$ -carotene concentration in plasma and a lower intake compared to young ones. With those results we can consider that serum levels of  $\beta$ -carotene are influenced by its intake.

Younger men have a higher  $\beta$ -carotene intake than the other groups. This young group had a higher vegetable intake which can explain the higher  $\beta$ -carotene level. Young men also have a higher plasma  $\beta$ -carotene concentration, compared to old men and a little higher than young women.

According to Morinobu et al (1994), the ratio of  $\beta$ -carotene to total lipids did not differ between elderly men and women. These authors considered that the difference in plasma  $\beta$ -carotene levels between elderly men and women was probably due to the differences in plasma lipids. In our study there is no correlation between plasma  $\beta$ -carotene and total lipids. We found positive correlations in the whole group, old people and women between plasma  $\beta$ -carotene and total cholesterol. In our subjects, old women have a higher  $\beta$ -carotene concentration in plasma and a higher plasma total cholesterol ( $p < 0.001$ ), compared to old men. No negative correlations between plasma  $\beta$ -carotene and triglycerides were found for the whole group, old people, women and old women. However, old women compared to men



have a higher  $\beta$ -carotene concentration but there is no difference in plasma triglycerides, between these two groups. With these results we can speculate that plasma  $\beta$ -carotene is influenced by plasma total cholesterol.

Comparing  $\beta$ -carotene concentration in plasma with the VERA-study reference values, we notice that only a small percentage of subjects (5.3 % of old men and 2.9 % of old women) have a concentration below the reference value of 10  $\mu\text{g}/\text{dl}$ . No subjects in the young group have a  $\beta$ -carotene concentration below the reference value (Table 22 appendix). These results indicate that the  $\beta$ -carotene status of our subjects is good.

#### 4.4 Selenium

Old people have a higher selenium plasma concentration than young. This higher concentration in the old group is a result of the difference found between old women and young, as men, old and young, have a similar serum selenium (Table 22). Most investigators (Campbell et al 1989, McAlpine et al 1989, Thomson et al 1977, Verlinden et al 1983, Lloyd et al 1983) agree that plasma levels of selenium decline with age. However our results are not in agreement with these studies.

Monget et al (1996) in their study about micronutrient status in elderly people observed 756 elderly people (193 men and 563 women), aged between 66 and 103 years, a little older than our subjects. These authors considered that the lower selenium plasma concentration in old people is due to a reduced intake of protein, since there is a significant correlation between the dietary intake of protein and serum selenium in the elderly. In our study, correlation between selenium plasma and protein intake is significantly positive in the whole group, in the young people of both genders and in women. Old women compared to young, have a higher percentage of total energy intake from protein and also a higher protein intake in grams per day. Correlation between serum selenium and protein intake is significantly positive for the young people. These results show that the lower protein intake can be the reason for the lower selenium plasma in young women.

Correlation between serum selenium and protein intake is significantly negative for the elderly people. Old women compared to old men, have a lower protein intake in grams per day ( $p < 0.05$ ).

According to Cabré et al (1992) serum selenium was directly associated with the percentage

of essential fatty acids and n-6 polyunsaturated fatty acids, and inversely related to percentage of saturated fatty acids in phospholipids. In our subjects correlation between serum selenium and total lipids is significantly positive for the whole group and for women. Old women showed a higher total plasma lipids than young ones and than old men ( $p < 0.001$ ). This may indicate that the higher selenium concentration observed in older women is due to higher plasma lipids.

When our results are compared to the VERA reference values, the younger subjects were found to have a higher proportion below the reference values (15.6% of young women and 12.5% of young men) than the older ones (7.8% of old women and 8.8% of old men) (Table 23 appendix).

## 5 Conclusion

In general, the antioxidant intake and serum levels seem to be acceptable in this population, with the exception of old men, in which group, 26.3% of the subjects have vitamin E intake below the reference value.

Although this study was carried out on a relatively small number of subjects, especially the younger men, the results suggest that, between old and young people, there are more differences on antioxidants plasma concentration than in their intake. The results suggest larger age differences in the antioxidants plasma concentrations than in their intake. The different antioxidant concentration, seems to be associated to ageing, as vitamin C, or to different intakes, as seems for  $\beta$ -carotene.

A decrease in immuno competence and a higher percentage of body fat, could explain the lower vitamin C concentration in old people, compared to the young ones, despite similar intakes. In relation to vitamin E, its concentration is strongly associated to plasma total lipids. Old people have higher vitamin E concentration in plasma, in comparison to young people, due to higher total lipids in blood.  $\beta$ -carotene concentration in plasma, seems to be influenced by  $\beta$ -carotene intake. Young men have a higher intake as well as a higher  $\beta$ -carotene concentration in plasma compared to old men. Association between plasma  $\beta$ -carotene and blood lipids was not very clear. We found different correlations between  $\beta$ -carotene plasma and total lipids, total cholesterol and triglycerides, that not confirm this association. Old women have a higher selenium concentration in plasma than young ones and old men. A higher protein intake (in grams and percentage) and a higher blood lipid level account for this higher concentration.



## 6 Summary

Data from the longitudinal study *Giessener Senioren Langzeitstudie* (GISELA-study) of the year 1998 (160 elderly people) and from the Nutrition and health examination study of young adults (NHESYA) (40 young people) were analyzed to compare antioxidant nutrient intake and plasma antioxidant levels on these two different age groups. Smokers and users of supplements as well as people who did not record their food intake were excluded.

Old people have lower plasma vitamin C despite a similar intake in comparison to young people.

After adjusting, vitamin E for total lipids, results showed that higher vitamin E plasma concentration in old people, despite a lower intake, is only a result of a higher blood lipids concentration.

Young men have a higher  $\beta$ -carotene concentration in plasma as well as a higher intake compared to old men. Old women have a higher  $\beta$ -carotene concentration than old men, despite not having a higher intake.

Old women have a higher selenium concentration in plasma, in comparison to young ones. Men, old and young, have a similar selenium concentration in plasma.

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## 8 Appendix

**Table 1:** Vitamin C concentration in plasma (mg/dl) in old and young people of VERA-study  
(Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	1.45	1.44
Men (n = 592)	1.07	1.28

**Table 2:** Vitamin C concentration in plasma (mg/dl) in old and young people  
(Mean)

	Old people (n = 156)	Young people (n = 40)
Women (n = 132)	1.27	1.50
Men (n = 64)	1.11	1.31

**Table 3:** Vitamin C intake (mg/d) in old and young people of VERA-study  
(Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	108	103
Men (n = 592)	87	101



**Table 4:** Vitamin C intake (mg/d) in old and young people  
(Mean)

	Old people (n = 160)	Young people (n = 40)
Women (n = 135)	111	109
Men (n = 65)	106	117

**Table 5:** Old and young people with vitamin C concentration in plasma below,  
VERA-study reference values  
(Percentage)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	2.9	0.0	2.2
Men (n = 64)	7.0	0.0	6.2
Whole group (n = 196)	4.4	0.0	3.5

**Table 6:** Old and young people with vitamin C intake below,  
RDAs reference values  
(Percentage)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 200)
Women (n = 135)	12.6	15.6	13.3
Men (n = 65)	12.3	25.0	13.8
Whole group (n = 200)	12.5	17.5	13.5

**Table 7:** Correlation between vitamin E concentration in plasma (mg/dl) and triglycerides concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = 0.379 p < 0.001	R = 0.594 p < 0.001	R = 0.505 p < 0.001
Men (n = 64)	R = 0.305 p < 0.05	R = 0.731 p < 0.05	R = 0.389 p < 0.01
Whole group (n = 195)	R = 0.373 p < 0.001	R = 0.600 p < 0.001	R = 0.477 p < 0.001

**Table 8:** Correlation between vitamin E concentration in plasma (mg/dl) and cholesterol concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = 0.491 p < 0.001	R = 0.854 p < 0.001	R = 0.674 p < 0.001
Men (n = 64)	R = 0.673 p < 0.001	R = 0.309 p = n.s.	R = 0.698 p < 0.001
Whole group (n = 195)	R = 0.597 p < 0.001	R = 0.775 p < 0.001	R = 0.703 p < 0.001

**Table 9:** Correlation between vitamin E concentration in plasma (mg/dl) and HDL cholesterol concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = - 0.059 p = n.s.	R = 0.062 p = n.s.	R = - 0.031 p = n.s.
Men (n = 64)	R = 0.175 p = n.s.	R = - 0.711 p < 0.05	R = 0.144 p = n.s.
Whole group (n = 195)	R = 0.077 p = n.s.	R = 0.016 p = n.s.	R = 0.055 p = n.s.

**Table 10:** Correlation between vitamin E concentration in plasma (mg/dl) and LDL cholesterol concentration in plasma (mg/dl) in old and young people (Spearman)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	R = 0.455 p < 0.001	R = 0.696 p < 0.001	R = 0.618 p < 0.001
Men (n = 64)	R = 0.634 p < 0.001	R = 0.319 p = n.s.	R = 0.661 p < 0.001
Whole group (n = 195)	R = 0.543 p < 0.001	R = 0.661 p < 0.001	R = 0.644 p < 0.001

**Table 11:** Vitamin E concentration in plasma (mg/dl) in old and young people of VERA-study (Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	1.56	1.15
Men (n = 592)	1.49	1.18



**Table 12:** Vitamin E concentration in plasma (mg/dl) in old and young people  
(Mean)

	Old people (n = 155)	Young people (n = 40)
Women (n = 131)	1.41	1.05
Men (n = 64)	1.24	1.09

**Table 13:** Vitamin E intake (mg/d) in old and young people of VERA-study  
(Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	14.3	13.4
Men (n = 592)	16.0	17.2

**Table 14:** Vitamin E intake (mg/d) in old and young people  
(Mean)

	Old people (n = 160)	Young people (n = 40)
Women (n = 135)	12.4	13.3
Men (n = 65)	12.6	17.1

**Table 15:** Old and young people with vitamin E concentration in plasma below,  
VERA-study reference values  
(Percentage)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	0.0	3.1	0.7
Men (n = 64)	1.8	0.0	1.5
Whole group (n = 195)	0.6	2.5	1.0

**Table 16:** Old and young people with vitamin E intake below,  
comparing with RDAs reference values  
(Percentage)

	Old people (n = 160)	Young people (n = 40)	Whole group (n = 200)
Women (n = 135)	18.4	12.5	17
Men (n = 65)	26.3	0.0	23.1

**Table 17:** Correlation between  $\beta$ -Carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) and  
total lipids concentration in plasma ( $\text{mg}/\text{dl}$ ) in old and young people  
(Spearman)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	R = 0.070 p = n.s.	R = - 0.087 p = n.s.	R = 0.005 p = n.s.
Men (n = 64)	R = - 0.096 p = n.s.	R = 0.143 p = n.s.	R = - 0.185 p = n.s.
Whole group (n = 196)	R = 0.093 p = n.s.	R = - 0.082 p = n.s.	R = - 0.016 p = n.s.

**Table 18:**  $\beta$ -Carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) in old and young people of VERA-study  
(Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	45	38
Men (n = 592)	30	30

**Table 19:**  $\beta$ -Carotene concentration in plasma ( $\mu\text{g}/\text{dl}$ ) in old and young people  
(Mean)

	Old people (n = 156)	Young people (n = 40)
Women (n = 132)	47	51
Men (n = 64)	34	62

**Table 20:**  $\beta$ -Carotene intake ( $\text{mg}/\text{d}$ ) in old and young people of VERA-study  
(Mean)

	Old people (n = 412)	Young people (n = 954)
Women (n = 774)	2.41	2.07
Men (n = 592)	2.19	2.00



**Table 21:**  $\beta$ -Carotene intake (mg/d) in old and young people  
(Mean)

	Old people (n = 160)	Young people (n = 40)
Women (n = 135)	3.60	5.36
Men (n = 65)	3.66	8.74

**Table 22:** Old and young people with  $\beta$ -carotene concentration in plasma below,  
comparing with VERA-study reference values  
(Percentage)

	Old people (n = 156)	Young people (n = 40)	Whole group (n = 196)
Women (n = 132)	2.9	0.0	2.2
Men (n = 64)	5.3	0.0	4.6
Whole group (n = 196)	3.8	0.0	3.0

**Table 23:** Old and young people with selenium concentration in plasma below,  
comparing VERA-study reference values  
(Percentage)

	Old people (n = 155)	Young people (n = 40)	Whole group (n = 195)
Women (n = 131)	7.8	15.6	9.6
Men (n = 64)	8.8	12.5	9.2
Whole group (n = 195)	8.1	15	9.5

**Table 24:** Classification of food items listed in the food record into food groups

Food groups	Food items
Bread, buns and cakes	mixed-grain bread/mixed roll, dark-/wholemeal bread/wholemeal roll, white bread/toast, water roll, swedish bread/biscuit, raisin roll/bun, scrambled eggs cake, yeast cake/biscuit, fruit cake, cream cake, biscuits
Pasta, rice, muesli, cornflakes	cerealfakes/muesli, cornflakes, rice, pasta , breadcrumbs
Milk and dairy products	milk until 1,5 % fat/buttermilk, milk until 3,5 % fat, evaporated milk, cacao drink, yoghurt natur/kefir/soured milk until 1,5 % fat, yoghurt natur/kefir/soured milk 3,5 % fat, fruitjoghurt until 1,5 % fat, fruitjoghurt 3,5 % fat, quark slim/coffage cheese, quark fat, fruitquark, cream fraiche, cheese until 40 % fat, cheese above 40 % fat
Fruits and fruit products	appel/pear/peach/nectarine, banana, orange, grapefruit, mandarine/zitrone, plum/morello cherries, grapes/berriesfruit, apricot, melon, kiwi, fruitpreserve/tinned fruit, dried fruit
Vegetables	peas/green bean, lentiles/fat bean, cabbage, sharp cabbage, tomate, cucumber, salad, spinach/leek, onion raw, onion cooked, mushroom, paprika raw, paprika cooked, carrot raw, carrot cooked, sweetcorn, courgette, radish, sharps vegetables, garlic, avocado
Potatos	potatos, chips/crisps, masched potato, roast potato, potato punch, potato ball/-dumpling
Meat and meat products	chops/steak, roast/meat cooked, mince, meatballs, salami/sausage/chorizo, stew, breeding/livestock farming, wild, liver/kidney/heart, ice leg, patés, streaky bacon/tripe, cured sausage slim, cured sausage fat, ham cooked, smoked ham
Fish	fish, fat fish, fish finget, fish preserve, prawn
Spread and cooking fat	butter/lard, margarine, half fat margarine, cooking fat

Sugar and sweets	jam/quince jelly, honey/syrup, nuts-nougat-cream, addition sugar/sweetener, pudding, ice, chocolate, sweets, muesli row, chewing gum/liquotice
Non-alcoholic drinks	tea/coffee, fruit tea/medicine herbs tea, mineralwater, fruitjuice, fruitnectar, multivitaminjuice, vegetablesjuice, limonade, lightdrink, beer without alcohol
Alcoholic drinks	beer, appelwine, wine/champagne, liqueur, witty drinks



## Brot, Brötchen

	So	Mo	Di
Mischbrot	Scheibe	45 g	
Mischbrötchen	Stück		
Schwarz-, Vollkornbrot	Scheibe	50 g	
Vollkornbrötchen	Stück		
Weißbrot, Toast	Scheibe	20 g	
Wasserbrötchen	Stück	45 g	
Knäckebröt	Scheibe	10 g	
Zwieback	Stück		
Rosinenbrötchen, Butterhörnchen	Stück	45 g	

## Süße Brotaufstriche

	So	Mo	Di
Konfitüre, Marmelade	Teelöffel	10 g	
Honig, Sirup	Teelöffel	10 g	
Nuß-Nougat-Creme	Teelöffel	10 g	

## Fette, Öle

	So	Mo	Di
Butter, Schmalz	Teelöffel	4 g	
Margarine	Teelöffel	4 g	
Halbfettmargarine	Teelöffel	4 g	
Speiseöl	Eßlöffel	10 g	

## Milch, Milchprodukte, Eier

	So	Mo	Di
Milch			
- bis 1,5 % Fett, Buttermilch	Tasse	150 ml	
- 3,5 % Fett	Tasse	150 ml	
Kondensmilch	Teelöffel	5 g	
Kakaogetränk	Tasse	150 ml	
Joghurt natur, Kefir, Dickmilch	Becher	150 g	
- bis 1,5 % Fett			
- 3,5 % Fett	Becher	150 g	
Fruchtojoghurt	Becher	150 g	
- bis 1,5 % Fett			
- 3,5 % Fett	Becher	150 g	
Quark			
- mager (bis 20 % Fett) Hüttenkäse	Eßlöffel	30 g	
- fett (über 20 % Fett)	Eßlöffel	30 g	
Fruchtquark	Eßlöffel	30 g	
Crème fraîche, Schmand	Eßlöffel	20 g	
Saure Sahne	Eßlöffel	20 g	
Schlagsahne	Eßlöffel	15 g	
Käse			
- bis 40 % Fett z.B. Harzer, Schichtkäse, fettreduzierter Käse	Scheibe/ Eßlöffel	30 g	
- über 40 % Fett z.B. Gouda, Edamer, Brie, Camembert	Scheibe/ Eßlöffel	30 g	
Ei	Stück	60 g	

2

1. Welche Mahlzeiten nimmst Du gewöhnlich im Laufe des Tages zu Dir?

- Erstes Frühstück  
 Zweites Frühstück  
 Mittagessen  
 Nachmittagsmahlzeit  
 Abendmahlzeit  
 Spätmahlzeit

2. Wie oft nimmst Du normalerweise eine warme Mahlzeit zu Dir?

- 1 - 3 mal pro Woche  
 4 - 6 mal pro Woche  
 7 mal pro Woche  
 8 - 10 mal pro Woche  
 11 mal pro Woche und mehr

3. Wer kocht bei Dir gewöhnlich?

(Es können auch mehrere Antworten angekreuzt werden.)

- Ich selbst  
 Mein Partner  
 Andere Personen, die in meiner Wohnung leben  
 Verwandte/Bekannte, die nicht in meiner Wohnung leben  
 Restaurant/Gaststätte  
 Sonstige, und zwar: \_\_\_\_\_  
 Mensa/Kantine

4. Wie wichtig sind Dir folgende Punkte beim Essen bzw. bei der Essenszubereitung?

	sehr wichtig	wichtig	es geht	weniger wichtig	unwichtig
Geschmack	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Preis	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Gesundheit	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Arbeits-/Zeitaufwand	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Bekömmlichkeit	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Gewohnheit	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>

5. Wieviele Tassen koffeinhaltigen Kaffees trinkst Du normalerweise am Tag?

- ich trinke keinen koffeinhaltigen Kaffee  
 weniger als 1 Tasse  
 1 - 2 Tassen  
 3 - 4 Tassen  
 5 Tassen und mehr

6. Wieviele Tassen schwarzen Tees trinkst Du normalerweise am Tag?

- ich trinke keinen schwarzen Tee  
 weniger als 1 Tasse  
 1 - 2 Tassen  
 3 - 4 Tassen  
 5 Tassen und mehr