Lifestyle and cardiovascular disease risk factors as determinants of total cysteine in plasma: the Hordaland Homocysteine Study[1–3]

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ABSTRACT

Background: Plasma total homocysteine (tHcy) is a cardiovascular disease risk factor and is related to several components of the established cardiovascular disease risk profile. Cysteine is structurally and metabolically related to homocysteine, but data on its association with cardiovascular disease and cardiovascular disease risk factors are sparse.

Objective: Our objective was to search for the determinants of plasma total cysteine (tCys) and compare them with those of tHcy.

Design: In this cross-sectional study, we studied 7591 healthy men and 8585 healthy women aged 40–67 y with no history of hypertension, diabetes mellitus, coronary heart disease, or cerebrovascular disease.

Results: In the group aged 40–42 y, tCys was significantly higher in men ($\bar{x}$: 273 μmol/L; 2.5–97.5 percentile: 219–338 μmol/L) than in women (253 μmol/L; 202–317 μmol/L) ($P < 0.001$). In the group aged 65–67 y, there was no significant sex difference in tCys: men (296 μmol/L; 233–362 μmol/L) and women (296 μmol/L; 234–361 μmol/L). As with tHcy, tCys was positively associated with age, total cholesterol concentration, diastolic blood pressure, and coffee consumption. Body mass index was a strong determinant of tCys but was not related to tHcy. Several factors known to influence tHcy, including smoking status, folate and vitamin intake, heart rate, and physical activity, were not associated or were only weakly associated with tCys.

Conclusion: Plasma tCys is strongly related to several factors that constitute the cardiovascular disease risk profile. This should be an incentive to determine the role of tCys in cardiovascular disease. *Am J Clin Nutr* 1999;70:1016–24.

KEY WORDS Plasma total cysteine, homocysteine, body mass index, total cholesterol concentration, coffee consumption, blood pressure, humans, cardiovascular disease risk factors, lifestyle, Hordaland Homocysteine Study, Norway

INTRODUCTION

About 100 epidemiologic and clinical studies that included >10 000 case subjects and an equal number of control subjects have shown that elevated plasma total homocysteine (tHcy) is an independent risk factor for occlusive disease in the coronary, cerebrovascular, and peripheral vessels and for arterial and venous thrombosis (1–5).

Homocysteine is formed from the essential amino acid methionine as a product of numerous *S*-adenosylmethionine–dependent transmethylation reactions (6). Homocysteine may be remethylated to methionine. This reaction is in most tissues catalyzed by the ubiquitous enzyme 5-methyltetrahydrofolate–homocysteine *S*-methyltransferase (methionine synthase), which requires vitamin B-12 as a cofactor and methyltetrahydrofolate as a cosubstrate (6). Alternatively, homocysteine is degraded to cysteine via the transsulfuration pathway in 2 sequential vitamin B-6–dependent reactions (6). Inhibition of homocysteine metabolism as a result of enzymatic defects or vitamin deficiencies causes homocysteine export into extracellular compartments and thereby causes elevated plasma tHcy (7).

Despite the strong evidence that tHcy is an independent risk factor for cardiovascular disease (5), the question of causality and the mechanism or mechanisms by which tHcy exerts its pathogenicity are still not clear. Experimental evidence suggests that the mechanism may be related to the reactivity or redox properties of the sulfhydryl group (8, 9). If this is the case, other thiols might also be expected to confer increased cardiovascular disease risk. Total cysteine (tCys), like homocysteine, is an aminothiol, and its concentration in plasma is ≈20-fold higher than tHcy (≈250 μmol/L) (10). However, there are only 2 reports showing higher tCys concentrations in vascular patients than in healthy control subjects (11, 12). In vascular disease (11) and in conditions characterized by transient (13, 14) or long-term (15, 16) elevation of tHcy, hyperhomocysteinemia was associated with complex changes in tCys and overall aminothiol redox status. Thus, hyperhomocysteinemia should not be considered an isolated event, but rather as a component of an interactive redox thiol system (10), the impairment of which may cause vascular lesions.

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In this study we investigated the possible relations between tCys and several demographic and lifestyle factors associated with increased cardiovascular disease risk. We used baseline tCys data from the Hordaland Homocysteine Study of 16176 healthy women and men (17). In this population, we showed that tHcy increases with age, is higher in men than in women, and is associated with risk factors such as smoking, high blood pressure, high blood cholesterol concentrations, and low physical activity. In addition, plasma tHcy is increased by high coffee consumption and is reduced by intake of folate and vitamins (18). A similar investigation of tCys may shed light on the possible role of tCys and tHcy as mediators of cardiovascular disease risk.

SUBJECTS AND METHODS

Study population

From April 1992 to April 1993, the National Health Screening Services, in cooperation with the University of Bergen and local health services, conducted the Hordaland Homocysteine Study in the Hordaland county of western Norway. A total of 24815 women and men from 3 age groups were invited to participate in the study. The youngest age group contained all subjects in the county who were 40–42 y of age. The oldest age group contained all subjects in Bergen and 3 neighboring suburban municipalities aged 65–67 y. The middle group, aged 43–64 y, was a 2% random sample of the residents in the city of Bergen. The attendance rate for the whole group was 72.7%, constituting a total of 18043 subjects.

To allow for any effect on tCys of disease, treatment, or change in lifestyle, 1866 participants who reported a previous diagnosis of coronary heart disease, cerebrovascular disease, hypertension, or diabetes mellitus were excluded from the analyses. One patient with homocystinuria was also excluded. Thus, 16176 subjects were included in this study. Because 96.2% of the participants belonged to the youngest and oldest age groups, most of the analyses were confined to these 2 groups. The study protocol was approved by the Regional Ethical Committee of western Norway, whose directives are based on the Helsinki Declaration.

Data collection

Data were collected via questionnaires, examinations, and blood tests. The questionnaires provided information about age; smoking habits; physical activity; type of work; personal history of cardiovascular diseases, hypertension, and diabetes; family history of disease; recent food intake; lifestyle; medical history; dietary habits; use of alcohol; and frequency of intake of various food items and vitamin supplements. Details on the collection and categorization of data were published previously (17).

Physical activity was categorized as *1*) sedentary or no activity, *2*) moderate activity (walking, cycling, or other type of moderate activity for ≥4 h/wk), *3*) active exercise (exercise, gardening with physical exertion, or a similar degree of activity for ≥4 h/wk, or *4*) heavy training (regular heavy training or participation in competitive sports several times a week). Subjects were classified into 5 categories: never smokers, former smokers, light smokers (1–9 cigarettes/d), moderate smokers (10–19 cigarettes/d), and heavy smokers (≥20 cigarettes/d). Coffee consumption was divided into 5 categories according to the number of cups consumed per day: 0, <1, 1–4, 5–8, and ≥9.

A vitamin-supplement score was created on the basis of use during the year and frequency of intake during the week of any type of vitamin supplement and was divided into 5 categories. The lowest category consisted of subjects who never used vitamin supplements and the highest category consisted of those who took vitamins 6–7 d/wk during the whole year (18). A folate score was computed from food-frequency information and data on the use of vitamin supplements. The calculation of the folate score and its validation against plasma folate and tHcy concentrations in 329 healthy subjects were reported previously (18).

Clinical examinations, blood sample collection, and biochemical analysis

The examinations included measurements of height, weight, and blood pressure; details of the procedures were reported previously (17). Blood was drawn from nonfasting subjects. Procedures for blood sample collection, processing, transport, and storage were described previously (17). Plasma tCys and tHcy were determined by HPLC and fluorescence detection (19, 20). The precision (between-day CV) of the assay is <3%. Serum total cholesterol and triacylglycerol concentrations were measured with enzymatic methods at the Department of Clinical Chemistry, Ullevål Hospital, Oslo.

Statistical methods

Because the distribution of tCys was symmetrical, no transformations were performed; arithmetic means are given for tCys. The distribution of tHcy was skewed with a long tail toward high values, and therefore the analyses were done by using $\log_{10}$ tHcy values.

Multiple linear regression models were used to assess the simultaneous relation among the various predictors of tCys. Plasma tCys was the dependent variable, whereas the independent variables were represented in the models as indicator variables denoting membership to 1 of 4 categories for cholesterol, heart rate, diastolic blood pressure, body mass index (BMI), physical activity, triacylglycerols, and folate intake and membership to 1 of 5 categories for smoking status, coffee consumption, and vitamin intake. Thus, the regression coefficient was used to estimate the difference in mean tCys between the reference category and the other categories for each factor. tCys concentrations across categories of each risk factor were tested jointly for homogeneity of means and for linear trend.

The multiple regression technique determines the relation between a set of independent variables and the mean of a normally distributed dependent variable (tCys). Thus, this model implies that the independent variables have the same effect on high and low values of the dependent variable. Biologically, however, it is possible that a factor influences extreme high or low values of the dependent variable with only moderate effects on the overall mean (21). Such effects can be overlooked with linear regression but may be studied with a set of logistic regression analyses, each at a different cutoff for the dichotomous dependent variable. Therefore, a series of logistic regressions were performed to assess the effect of the different factors on high and low tCys concentrations. The 5th (tCys ≤ 217 μmol/L) and 95th (tCys ≥ 330 μmol/L) percentiles of the total study sample were chosen as cutoffs. The results, presented as odds ratios, represented an estimate of the risk that an individual with a specific risk profile was hypo- or hypercysteinemic relative to subjects with a baseline risk profile.

Furthermore, Pearson correlation coefficients were computed to provide a simpler summary of the linear relations between tCys and the various factors. The analyses were performed by

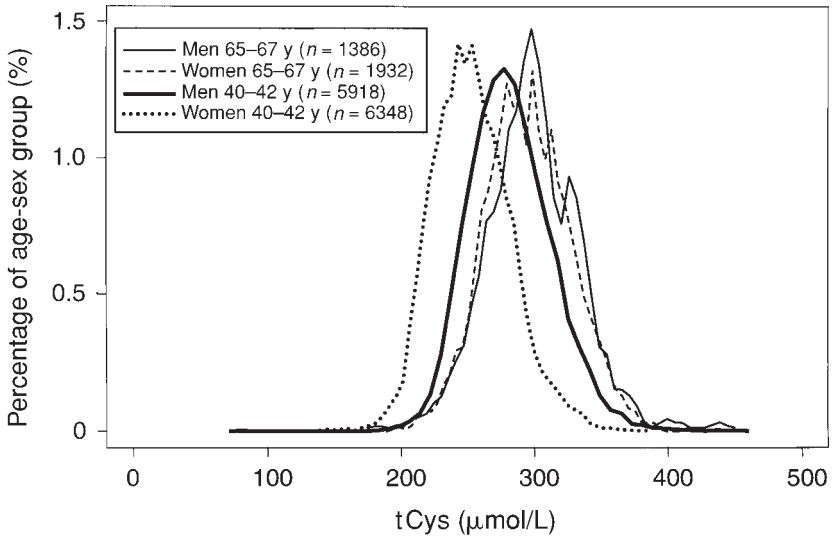

FIGURE 1. Distribution of plasma total cysteine (tCys) in the 4 main age and sex groups.

using the statistical package BMDP (22). In addition, S-PLUS software (23) was used to construct Lowess' plots (24) of the smoothed relations between 2 variables and to estimate the density distribution of tCys. All tests were two-tailed, and a P value <0.05 was considered significant.

RESULTS

Plasma tCys by age and sex

The distribution of tCys was symmetric. The concentrations ranged from 72.2 to 441.3 μmol/L, with overall mean and median values of 270.2 and 268.0 μmol/L, respectively. The distribution of tCys in the 4 main age and sex groups is shown in **Figure 1**. Mean tCys values and the 2.5 and 97.5 percentiles in the 3 age groups and in both men and women are shown in **Table 1**. Mean tCys values were higher in the older groups than in the youngest group. In the oldest age group there was no sex difference, whereas in the younger age groups tCys concentrations were higher in men than in women.

Relation of plasma tCys to cardiovascular disease risk and lifestyle factors

Predictors of tCys identified in a multiple regression analysis are shown in **Table 2**. For all variables, we estimated the difference in mean tCys concentrations between categories of each risk factor and its reference category. The results, adjusted for sex, age, or both, showed that the largest overall differences in tCys concentrations were observed across different categories of age, sex, BMI, and cholesterol concentration and, to a lesser extent, coffee consumption and diastolic blood pressure. Small but significant differences in tCys concentrations were found across different categories of smoking status, but there was no marked linear trend. Nonsignificant differences in tCys were observed for physical activity, heart rate, triacylglycerol concentration, and folate intake (data not shown). Additional adjustment for all factors in Table 2, in addition to folate score, heart rate, and physical activity, showed that the relations with age, BMI, cholesterol concentration, sex, coffee consumption, and diastolic blood pressure remained strong, whereas smoking status no longer contributed to the prediction of tCys concentrations.

To investigate whether the relations between tCys and the independent variables differed along the tCys distribution, we performed a series of logistic regressions with high and low plasma tCys as the outcome variables (**Table 3**). Age, sex, BMI, cholesterol concentration, diastolic blood pressure, and coffee consumption were associated with a shift of the tCys distribution to higher values. Although the associations with age and sex were somewhat stronger at low concentrations and stronger with BMI at high concentrations, the analyses of tail effects provided essentially the same information as the analyses of mean plasma tCys concentrations.

TABLE 1
Plasma total cysteine (tCys) concentration by age and sex

	40–42 y		43–64 y		65–67 y		
Sex	n	tCys	n	tCys	n	tCys	P^1
		μmol/L		μmol/L		μmol/L	
Male	5918	273.1 (218.6–338.4)[2]	287	279.5 (225.6–332.8)	1386	296.4 (232.9–362.2)	<0.001
Female	6348	253.1 (202.1–317.1)	305	275.8 (215.4–347.2)	1932	296.3 (233.5–360.5)	<0.001

[1] For linear trend.
[2] $\bar{x}$ (2.5–97.5 percentiles).

TABLE 2
Estimated change in plasma total cysteine (tCys) concentration by cardiovascular disease risk factor[1]

Risk factor	Estimated change in tCys		Estimated change in tCys	
	Adjusted for sex, age, or both	*P*	Multiple adjustment[2]	*P*
	μmol/L		μmol/L	
Sex				
Female (*n* = 8585)[3]				
Male (*n* = 7591)	15.35 (14.39, 16.31)	<0.001[4,5]	14.71 (13.28, 16.14)	<0.001[4,5]
Age (y)				
40–42 (*n* = 12 266)[3]				
43–64 (*n* = 592)	14.81 (12.26, 17.37)	<0.001[4,5]	9.64 (6.42, 12.87)	<0.001[4,5]
65–67 (*n* = 3318)	34.59 (33.40, 35.78)		29.56 (27.72, 31.40)	
BMI (kg/m^2)				
<20 (*n* = 873)[3]				
20–24.99 (*n* = 8358)	7.21 (5.10, 9.33)		6.01 (3.31, 8.72)	
25–29.99 (*n* = 5731)	17.29 (15.11, 19.48)	<0.001[4,5]	14.09 (11.25, 16.94)	<0.001[4,5]
≥30 (*n* = 1191)	29.00 (26.35, 31.65)		26.54 (23.01, 30.08)	
Cholesterol (mmol/L)				
<4.00 (*n* = 570)[3]				
4.00–5.99 (*n* = 9235)	9.58 (6.98, 12.18)		7.82 (4.65, 10.98)	
6.00–7.99 (*n* = 5627)	16.48 (13.80, 19.16)	<0.001[4,5]	12.92 (9.58, 16.26)	<0.001[4,5]
≥8.00 (*n* = 746)	21.99 (18.55, 25.42)		18.44 (13.84, 23.05)	
Diastolic blood pressure (mm Hg)				
<70 (*n* = 2583)[3]				
70–84 (*n* = 8571)	4.80 (3.43, 6.16)		2.92 (1.23, 4.60)	
85–99 (*n* = 4149)	9.75 (8.20, 11.30)	<0.001[4,5]	5.83 (4.10, 7.57)	<0.001[4,5]
≥100 (*n* = 847)	13.69 (11.24, 16.14)		8.27 (4.88, 11.67)	
Coffee (cups/d)				
0 (*n* = 1285)[3]				
<1 (*n* = 485)	6.29 (3.08, 9.51)		6.02 (2.09, 9.96)	
1–4 (*n* = 8146)	11.15 (9.33, 12.97)	<0.001[4,5]	10.46 (8.21, 12.70)	<0.001[4,5]
5–8 (*n* = 5286)	13.56 (11.68, 15.44)		14.42 (12.06, 16.79)	
≥9 (*n* = 974)	16.19 (13.61, 18.76)		14.90 (11.54, 18.26)	
Smoking status				
Never smoker (*n* = 6078)[3]				
Former smoker (*n* = 4140)	0.81 (−0.42, 2.05)		−1.12 (−2.65, 0.40)	
Light smoker (*n* = 1373)	−1.75 (−3.56, 0.07)	0.002[4]	−1.71 (−4.06, 0.64)	0.04[4]
Moderate smoker (*n* = 3228)	−1.66 (−2.99, −0.32)	0.13[5]	−2.72 (−4.47, −0.97)	0.00[5]
Heavy smoker (*n* = 1278)	1.15 (−0.75, 3.04)		−1.25 (−3.82, 1.32)	
Vitamin supplement score[6]				
Level 0 (*n* = 4066)[3]				
Level 1 (*n* = 1904)	0.05 (−1.65, 1.74)		−0.70 (−2.54, 1.15)	
Level 2 (*n* = 2675)	0.23 (−1.31, 1.77)	0.99[4]	−1.22 (−3.39, 0.96)	0.60[4]
Level 3 (*n* = 1685)	0.07 (−1.72, 1.87)	0.76[5]	−1.36 (−4.37, 1.65)	0.07[5]
Level 4 (*n* = 1612)	0.39 (−1.44, 2.22)		0.12 (−3.33, 3.56)	

[1] 95% CIs in parentheses.
[2] Adjusted for all risk factors in this table and for folate score, physical activity, and heart rate.
[3] Reference category.
[4] All levels of the risk factor were tested jointly (test for homogeneity).
[5] For linear trend.
[6] Level 0 included subjects who had never used vitamin supplements and level 4 included subjects who used supplements 6–7 d/wk during the whole year.

We also studied possible effect modifications by age and sex by investigating the correlations between tCys and the various factors in subgroups (**Table 4**). In all age and sex groups, plasma tCys concentrations correlated strongly with BMI (**Figure 2**) and also correlated with cholesterol concentration and diastolic blood pressure. The relation with BMI was attributed mainly to weight and not to height (data not shown). The association of tCys with coffee intake was strongest in the youngest age group. In contrast, smoking was negatively associated with tCys in the elderly. Weaker or no correlations were found between tCys and triacylglycerols, heart rate, physical activity, and vitamin intake (data not shown).

There was a strong nonlinear relation between tCys and tHcy. Up to a tHcy concentration of 12 μmol/L, there was a strong positive association between tCys and tHcy, but at higher tHcy concentrations the relation was inverse (**Figure 3**). To exclude the possibility that the associations between tCys and the various factors investigated were secondary to changes in tHcy, we repeated the correlation analyses after dividing the subjects into those with tHcy concentrations >12 μmol/L and those with tHcy concentrations ≤12 μmol/L. The association between tCys and the various factors (except for tHcy) remained essentially unaltered (Table 4).

TABLE 3
Odds ratio (OR) for hypo- and hypercysteinemia, adjusted for sex and age, by cardiovascular disease risk factor[1]

	tCys ≤ 217 μmol/L			tCys ≥ 330 μmol/L	
Risk factor	OR (95% CI)	1/OR	P^2	OR (95% CI)	P^2
Sex					
Female (n = 8585)[3]	1			1	
Male (n = 7591)	0.21 (0.17, 0.26)	4.68	<0.001	1.61 (1.41, 1.88)	<0.001
Age (y)					
40–42 (n = 12 266)[3]	1			1	
43–64 (n = 592)	0.29 (0.16, 0.54)	3.42	<0.001	2.11 (1.46, 3.07)	<0.001
65–67 (n = 3318)	0.09 (0.06, 0.14)	11.1		6.68 (5.76, 7.74)	
BMI (kg/m^2)					
<20 (n = 873)[3]	1			1	
20–24.99 (n = 8358)	0.70 (0.55, 0.89)	1.43		1.95 (1.11, 3.44)	
25–29.99 (n = 5731)	0.41 (0.31, 0.54)	2.44	<0.001	3.84 (2.18, 6.75)	<0.001
≥30 (n = 1191)	0.26 (0.17, 0.42)	3.82		9.03 (5.06, 16.1)	
Cholesterol (mmol/L)					
4.00 (n = 570)[3]	1			1	
4.00–5.99 (n = 9235)	0.57 (0.43, 0.76)	1.75		1.39 (0.75, 2.57)	
6.00–7.99 (n = 5627)	0.39 (0.28, 0.53)	2.60	<0.001	2.05 (1.11, 3.79)	<0.001
≥8.00 (n = 746)	0.42 (0.22, 0.79)	2.38		3.25 (1.71, 6.18)	
Diastolic blood pressure (mm Hg)					
<70 (n = 2583)[3]	1			1	
70–84 (n = 8571)	0.80 (0.67, 0.96)	1.24		1.48 (1.12, 1.95)	
85–99 (n = 4149)	0.60 (0.48, 0.77)	1.66	<0.001	2.19 (1.65, 2.90)	<0.001
≥100 (n = 847)	0.48 (0.28, 0.85)	2.07		2.75 (1.97, 3.83)	
Coffee, (cups/d)					
0 (n = 1285)[3]	1			1	
<1 (n = 485)	0.71 (0.45, 1.10)	1.42		1.70 (0.98, 2.97)	
1–4 (n = 8146)	0.59 (0.47, 0.73)	1.70		1.87 (1.27, 2.75)	
5–8 (n = 5286)	0.42 (0.33, 0.53)	2.40	<0.001	2.10 (1.41, 3.12)	<0.001
≥9 (n = 974)	0.42 (0.28, 0.64)	2.36		2.70 (1.69, 4.32)	

[1] tCys, plasma total cysteine concentration.
[2] For linear trend.
[3] Reference category.

Lifestyle and risk factors according to tCys and tHcy response

To summarize the results and to highlight the differences between tCys and tHcy as response indicators, we grouped the lifestyle and risk factors into 4 categories according to the combined relations with tCys and tHcy (**Figure 4**). Group 1 were factors associated with both tCys and tHcy: age, cholesterol concentration, diastolic blood pressure, and coffee consumption. BMI was strongly related to tCys but not to tHcy (group 2), whereas smoking status, folate and vitamin intakes, heart rate, and physical activity were mainly associated with tHcy and not with tCys (group 3). Finally, triacylglycerol concentrations were weakly associated with both aminothiols (group 4).

DISCUSSION

We investigated plasma tCys concentrations and their relation to lifestyle and cardiovascular disease risk factors among 16 176 healthy adults in the Hordaland Homocysteine Study. The strongest determinants of tCys were age, BMI, sex, diastolic blood pressure, serum cholesterol concentration, and coffee consumption. These factors, except for BMI, were also associated with tHcy, which in turn showed relations not shown for tCys (17, 25). On the basis of their relation with tCys and tHcy concentrations, these factors were divided into 4 groups, as depicted in Figure 4. Such classification may point to possible mechanisms behind the associations and to a possible role of tCys as a cardiovascular disease risk factor.

Age, sex, cholesterol concentration, diastolic blood pressure, and coffee consumption were strong determinants of both tHcy and tCys and therefore complied with the criteria of group 1. Like tHcy (17, 26), tCys (Table 1) increased with age in both men and women. As with tHcy (27), the effect was most pronounced at the lower part of the tCys distribution (Table 3). Possible mechanisms are an age-dependent decrease in enzymatic activities involved in both cysteine and homocysteine metabolism (28) and impaired renal function. The latter possibility is supported by consistent observations that tCys and tHcy increase considerably in renal insufficiency (29). Notably, the decline in glomerular filtration rate may explain the age-related increase in tCys and tHcy (30).

Some (31), but not all (32), previous studies showed higher tCys concentrations in men than in women; such sex differences have been consistently shown for tHcy (17, 26). In the present study, plasma tCys concentrations were higher in men than in women in the youngest but not in the older age groups. The sex difference may have been due to hormonal effects, which vanish at advanced ages. Higher creatine-creatinine synthesis (a function of muscle mass and the major source of Hcy formation) in men than in women may contribute to the sex difference in tHcy concentration (26) but may have less of an effect on cysteine homeostasis.

Both tCys and tHcy were related to cholesterol concentration and diastolic blood pressure, and the relations remained strong after adjustment for other risk factors (Table 2). We are aware of no

TABLE 4
Correlation coefficients for plasma total cysteine concentrations with cardiovascular disease risk factors in young and old women and men and in subjects with a plasma total homocysteine (tHcy) concentration ≤12 μmol/L (low) or >12 μmol/L (high)[1]

	40–42 y						65–67 y					
	Men			Women			Men			Women		
Risk factor	All (n = 5918)	Low (n = 4212)	High (n = 1706)	All (n = 6348)	Low (n = 5434)	High (n = 914)	All (n = 1386)	Low (n = 702)	High (n = 684)	All (n = 1932)	Low (n = 1282)	High (n = 650)
BMI	0.25	0.25	0.25	0.22	0.22	0.24	0.21	0.21	0.24	0.30	0.31	0.31
Cholesterol concentration	0.14	0.14	0.10	0.12	0.12	0.09	0.10	0.03[2]	0.15	0.05	0.05[2]	0.03[2]
Diastolic blood pressure	0.15	0.13	0.14	0.12	0.12	0.10	0.14	0.10	0.15	0.08	0.10	0.04[2]
Coffee consumption	0.11	0.09	0.06	0.15	0.13	0.12	0.07	0.07[2]	−0.01[2]	0.06	0.03[2]	0.05[2]
Smoking status	−0.02[2]	−0.05	−0.06	0.03	0.02[2]	−0.05[2]	−0.06	−0.08	−0.12	−0.07	−0.13	−0.11
tHcy	0.27	0.35	−0.13	0.33	0.40	−0.23	0.26	0.36	−0.04[2]	0.30	0.33	−0.06[2]

[1] All coefficients significant (P < 0.05) unless marked otherwise.
[2] NS.

experimental or clinical observations that explain the relation between plasma aminothiol and cholesterol concentrations. The association of both tCys and tHcy with blood pressure may be related to the fact that both compounds react with nitric oxide to form vasoactive nitrosothiol adducts (8). In addition, cysteine may exert endothelium-dependent contraction by generating O_2^-, which rapidly inactivates the endothelium-derived relaxing factor (33).

A moderately strong relation between tCys and coffee consumption was observed. A similar relation was seen with tHcy and was attributed to the possible influence of caffeine (27). Notably, coffee consumption was associated with a complete shift of the tCys distribution (Table 3), but it was related to tHcy only at low concentrations (27).

BMI was a strong determinant of tCys in both men and women and in both age groups and was particularly predictive of high tCys concentrations. BMI has only rarely been associated with tHcy concentrations (34); in the present study we found no association and, therefore, BMI was classified as a group 2 factor. The mechanism for the association between BMI and tCys is unclear, but the association may reflect a possible role of plasma cysteine availability in stabilizing body cell mass (35).

Smoking status, vitamin and folate intakes, physical activity, and heart rate are determinants of plasma tHcy concentrations (17) but were essentially unrelated to tCys (Figure 4). Accordingly, these factors were classified as group 3 factors. However, the differential effects were not absolute because tCys was weakly related to heart rate and weakly and inversely related to smoking in elderly men and women. In young men and women, tCys was unrelated to smoking. In line with these findings, a recent study showed no effect of smoking on various cysteine forms in plasma (32). On the contrary, plasma tHcy showed a significant relation with smoking status that may have been attributable to changes in the thiol redox status or to the lower concentrations of plasma folate and vitamin B-12 in smokers (36).

As expected, tCys was unrelated to the intake of vitamin supplements or to the folate score, both of which are strong determinants of tHcy (Figure 4) (17, 18). Vitamin B-6, which is a cofactor in the 2 sequential enzymes that convert homocysteine into cysteine (6), was not assessed in the present study but should be considered as a possible factor for explaining some of the relations between tCys and several lifestyle factors. For instance, the strong relation between tHcy and coffee consumption (27) may have been due to impaired vitamin B-6 function, especially if caffeine functions as a vitamin B-6 antagonist, as shown recently for another xanthine derivative, theophylline (37). However, inhibition of the transsulfuration pathway is expected to decrease tCys, whereas we found that tCys increased in coffee drinkers. Smoking may also impair vitamin B-6 status, but elevated tHcy (38) and low tCys (Table 4) concentrations were observed only in the elderly smokers. Thus, our data do not support the hypothesis that coffee and smoking affect tCys by interfering with vitamin B-6 status. However, caffeine was shown recently to reduce the glomerular filtration rate (39), an effect that may partly have accounted for the increase in concentrations of both tCys and tHcy.

The present study has no clinical endpoints but addresses several questions of clinical relevance. Despite the relation

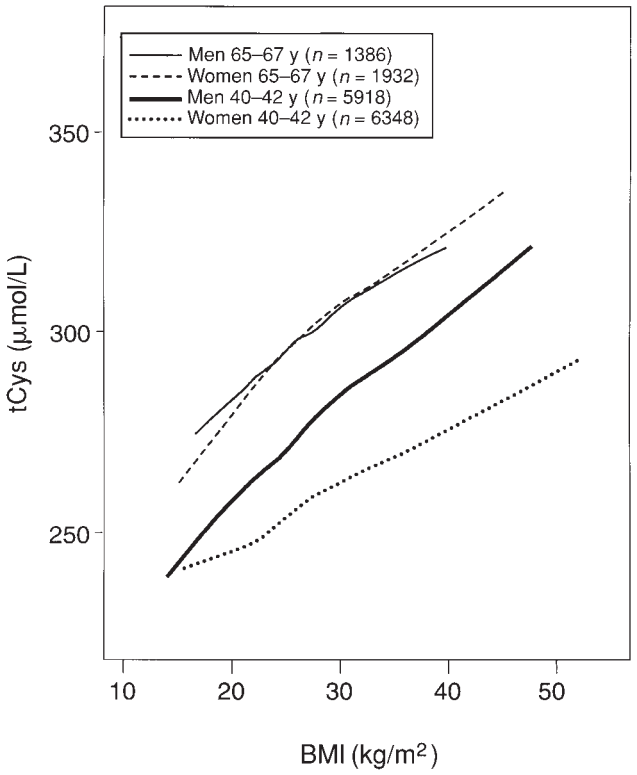

FIGURE 2. Relation between plasma total cysteine (tCys) and BMI in the 4 main age and sex groups.

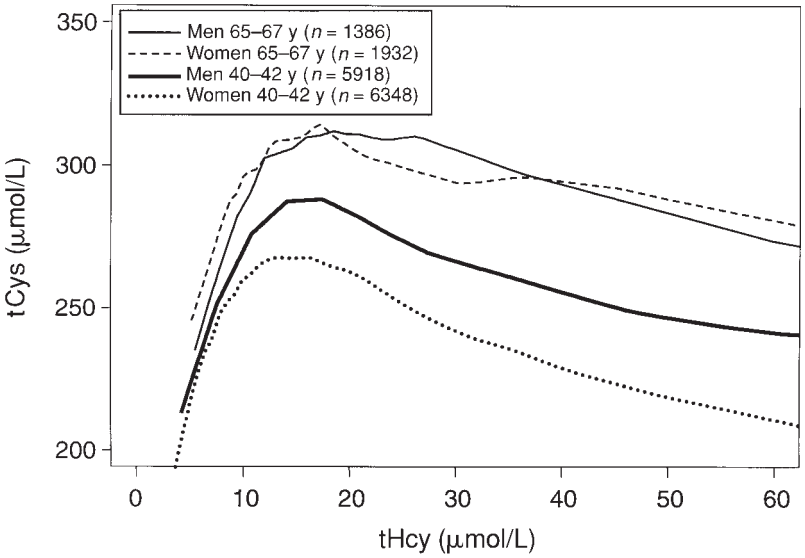

FIGURE 3. Relation between plasma total cysteine (tCys) and plasma total homocysteine (tHcy) in the 4 main age and sex groups.

between tCys and tHcy (Figure 3), the unique associations observed for each aminothiol (Figure 4) suggest that tCys and tHcy may confer independent cardiovascular disease risk. The possibility of an effect of tCys independent of tHcy is supported by similar relations between tCys and BMI, coffee consumption, serum cholesterol concentration, and blood pressure in subgroups with a high or low tHcy concentration (Table 4). Several in vitro studies on vascular or atherogenic effects of aminothiols showed similar effects for cysteine and homocysteine (5, 40). Such findings have been interpreted as a lack of specificity for homocysteine (5, 40) but could suggest that high concentrations of tCys and tHcy have a synergistic effect. Such synergy may explain the high cardiovascular mortality and morbidity predicted by hyperhomocysteinemia in patients with renal failure (41, 42), which, in contrast with vitamin deficiency, elevates both tHcy and tCys concentrations (43).

In conclusion, both tHcy and tCys are interactive components that undergo disulfide exchange and redox reactions (plasma redox thiol status) (10). Although tHcy has been established as an independent cardiovascular disease risk factor (5), only sparse data link tCys to human health and disease. Our study showed that tCys is strongly related to several biochemical, physical, and lifestyle factors, some of which also correlate with tHcy. Thus, these plasma aminothiols have distinct correlation profiles with established cardiovascular disease risk factors. Our data may suggest interactive effects in relation to the pathogenesis of vascular disease and should motivate clinical studies of these aminothiols as risk factors.

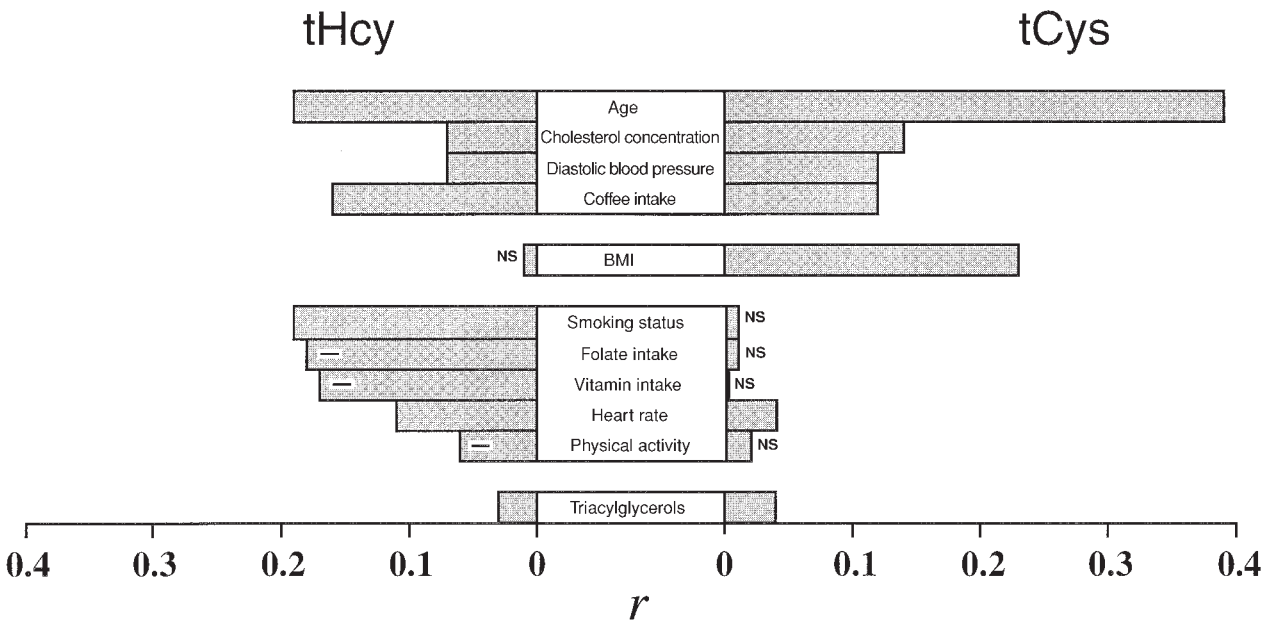

FIGURE 4. Relation between plasma total cysteine (tCys) and plasma total homocysteine (tHcy) and various cardiovascular disease risk factors. The minus sign indicates a negative correlation. $P > 0.05$.

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