Genetic Analysis of Indicators of Cholesterol Synthesis and Absorption: Lathosterol and Phytosterols in Dutch Twins and Their Parents

Dorret I. Boomsma[1], Hans M. Princen[2], Rune R. Frants[3], Jan A.Gevers Leuven[2], and Herman-Jan M. Kempen[2]

[1] *Department of Biological Psychology, Vrije Universiteit,*
[2] *Gaubius Laboratory PG-TNO, Leiden, The Netherlands*
[3] *Leiden University Medical Centre, Leiden, The Netherlands*

Significant familial aggregation was observed for plasma levels of lathosterol (an indicator of whole-body cholesterol synthesis) and plant sterols campesterol and β-sitosterol (indicators of cholesterol absorption) in 160 Dutch families consisting of adolescent mono- and dizygotic twin pairs and their parents. For lathosterol a moderate genetic heritability in parents and offspring (29%) was found. In addition, shared environment also contributed significantly (37%) to variation in plasma lathosterol concentrations in twin siblings. However, a model with different genetic heritabilities in the two generations (10% in parents and 68% in offspring) fitted the data almost as well. For plasma plant sterol concentrations high heritabilities were found. For campesterol heritability was 80% and for β-sitosterol it was 73%, without evidence for differences in heritability between sexes or generations. No influence of common environmental influences shared by family members was seen for either campesterol or β-sitosterol. Taken together, these results confirm and expand the hypothesis that individual differences in plasma levels of non-cholesterol sterols are moderately (lathosterol) to highly (plant sterols) heritable.

The rates of cholesterol synthesis and cholesterol absorption in man are of considerable scientific and clinical interest. Plasma levels of lathosterol are an indicator of whole-body cholesterol synthesis (Bjorkhem et al., 1987; Kempen et al., 1988; Miettinen, 1981; Miettinen et al., 1990). Plasma plant sterol concentrations reflect fractional cholesterol absorption (Tilvis & Miettinen, 1986). Plant sterols are present in most human diets (e.g., in fat-rich vegetables and fruits, nuts and vegetable oils) with around 65% as β-sitosterol and 30% as campesterol. Only a small fraction of plant sterols in the diet is absorbed (< 5%) and there is no evidence that plant sterols are synthesized by humans or animals (Salen et al., 1970). A rare familial disorder has been described — called phytosterolemia or sitosterolemia (Bhattacharyya & Connor, 1974) with elevated plant sterol levels in blood. Patients with the disorder may develop premature atherosclerosis (Beaty et al., 1986; Salen et al., 1983). It has been suggested that this disorder is inherited as an autosomal recessive trait (Bhattacharyya & Connor, 1974) against a background of significant familial correlation for β-sitosterol (Beaty et al., 1986). Glueck et al. (1991) observed in a sample of 16 hyperphytosterolemic probands and 34 of their first degree relatives a familial correlation of 0.2 for total serum phytosterol levels. The factors that are responsible for this familial resemblance might be polygenic or shared environmental influences, or a combination of both. Very little is known about the causes of familial resemblance for either phytosterol or lathosterol levels in the general population. One study by Berge et al. (2002) investigated the degree of genetic and environmental determination of variation between individuals in lathosterol and phytosterol levels. Regression of offspring sterol levels on midparental values suggested moderate to high heritabilities (42% for lathosterol, 81% for β-sitosterol and 84% for campesterol). These heritabilities were confirmed in a small sample of 12 MZ and 12 DZ twin pairs who also took part in the study. Correlations between MZ twins for lathosterol and phytosterol levels were substantially higher (between .63 and .88) than correlations between DZ twins (between .04 and .13).

In a previous paper (Kempen et al., 1991) we reported on the distribution of lathosterol, campesterol and β-sitosterol in a random sample of 160 Dutch families consisting of twins living with their parents, and the relationship of these sterols with body weight and plasma lipid levels. In the present paper we address the question to what extent inter-individual variation in these indicators of cholesterol metabolism is determined by genetic and environmental factors. To study the genetic and environmental contributions to variation in lathosterol, campesterol and β-sitosterol in the general population we measured these variables in monozygotic (MZ) and dizygotic (DZ) adolescent twin pairs and their parents. Studies of MZ and DZ

Address for correspondence: Dorret Boomsma, Dept Biological Psychology, Van der Boechorststraat 1, 1081 BT, Amsterdam, The Netherlands. Email: dorret@psy.vu.nl

twins provide a unique opportunity to separate common environmental variance shared by family members from genetic variance. If familial correlations are explained by genetic factors, MZ twins are expected to be at least twice as similar as DZ twins and parents and offspring, because MZ twins have identical genotypes. DZ twins share on average half of the additive gene effects and the correlation between their genotypes is .5. Genetic values of parents and offspring are also correlated .5, based on Mendelian inheritance. Under a purely shared environmental model MZ and DZ correlations are predicted to be the same, and parent-offspring correlations depend on the extent to which the family environment is shared by parents and their children. Studying male and female twins allows the assessment of whether the contributions of genetic and environmental factors differ in magnitude between the sexes and studying DZ twins of unlike sex makes it possible to test whether the genetic or environmental effects that are expressed in one sex differ from the effects expressed in the other sex. By including parents of twins the possible contribution of assortative mating and intergenerational differences in heritabilities can be assessed.

Subjects and Method

This study is part of a larger project in which cardiovascular risk factors were studied in adolescent twin pairs and their parents. Addresses of twins (between 14–21 years of age) living in Amsterdam and neighboring cities were obtained from City Council population registries. Twins living with both their biological parents were contacted by letter and asked to participate in the study. A family was included in the study if the twins and both parents were willing to participate. In addition, a small number of families who heard of the study from other twins volunteered to take part. Zygosity was determined by typing the following blood group polymorphisms: ABO, MNS, Rhesus, P, Lutheran, Kell, Duffy, Kidd, Gm, Am and Km and was confirmed by typing DNA polymorphisms (Beekman et al., 2002). Data from three series of triplets were included by discarding the data from the middle child. There were 35 MZ female pairs (average age 16.0, *SD* = 2.2), 35 MZ male pairs (16.6, *SD* = 1.8), 30 DZ female pairs (17.7, *SD* = 2.0), 31 DZ male pairs (17.2, *SD* = 1.7) and 29 DZ opposite sex pairs (16.4, *SD* = 1.8). Average age of fathers was 48.1 (*SD* = 6.3) and of mothers 45.6 (*SD* = 5.9) years. For 1 family with MZF twins no data were available for 1 child, data from 2 other families were not analyzed because children had extreme campesterol (DZM) or sitosterol (DOS) values. Data were not available for 2 fathers and 1 mother of twins, but data from their children were used in the genetic analyses. Fasting blood samples were taken between 8:30 and 10:30 a.m. by venipuncture, using Becton-Dickinson Vacutainers containing sodium-EDTA. Plasma was immediately separated from the cells and stored at –20° Celsius until further use. Concentrations of lathosterol and plant sterols were measured with gas chromatography (for details see Kempen et al., 1991).

Statistical Analysis

We (Kempen et al., 1991) found significant positive correlations between plasma lathosterol and body weight for fathers, mothers, sons and daughters (correlations between 0.2 and 0.3) and negative correlations between phytosterols and body weight (correlations between –0.1 and –0.3). All data were therefore corrected for body weight, separately in each generation by sex group. Plasma samples from family members were always measured in the same batch. A significant batch effect was observed (accounting for 8, 17 and 13% of the variance in lathosterol, campesterol and β-sitosterol, respectively). Before the genetic analyses were carried out, the data were therefore corrected for batch effects to exclude the possibility that familial resemblance would be influenced by inter-assay fluctuations. Data were logarithmically transformed, because the distribution of all 3 variables was skewed. For each trait, the corrected scores were summarized into 4 × 4 (Father, Mother, Twin 1, Twin 2) covariance matrices for each of the 5 sex by zygosity groups (i.e., families of MZ male and female twins, families of DZ male, female and opposite-sex twins). Likelihood ratio χ^2 tests were used to assess whether there are differences in variances between generations and between sexes by comparing the fit of a model which constrains variances to be equal across sexes or generations to one which allows them to vary, while taking into account that data from family members do not represent independent observations. The Chi-squared statistic is computed by subtracting the χ^2 for the full model from that for the reduced model. The degrees of freedom (*df*) for this test are equal to the difference between the df for the full and the reduced model (Neale & Cardon, 1992). Likewise, the effects of sex and zygosity on correlations between family members for lathosterol, campesterol and β-sitosterol were assessed by likelihood ratio χ^2 test. The effects of sex, generation and zygosity on variances of twins and parents, and on correlations between family members were examined with the LISREL7 computer program (Joreskog & Sorbom, 1988), which incorporates a maximum likelihood technique for parameter estimation.

Genetic models specified variation in observed phenotypes to be due to unobserved or latent genotypes and environments. These latent factors were G, additive genetic variation, C, common environmental variance shared by family members and E, environmental variation that is not shared by family members. The significance of these factors was tested by comparing the fit of, for example a GE model to that of the full GCE model. If the common environment is present, then the GE model will fit the data worse than the full model. Goodness-of-fit was assessed by likelihood ratio χ^2 tests. The overall χ^2 tests the agreement between the observed and the predicted variances and covariances in the 5 family groupings. A large χ^2 indicates a poor fit, while a small χ^2 indicates that the data are consistent with the model. Heritability was calculated from the estimated variance components as the genetic variance divided by the total phenotypic variance.

Results

Table 1 presents descriptive statistics and correlations between family members. In Table 1a the means of untransformed lathosterol, campesterol and β-sitosterol are listed

for fathers, mothers, sons and daughters. A significant sex difference between lathosterol concentrations of fathers and mothers was observed, as well as a significant generation difference between lathosterol concentrations of parents and offspring. There were no sex or generation differences in mean scores for campesterol and β-sitosterol (Kempen et al., 1991).

In Table 1b the standard deviations of the logarithmically transformed scores are given and Table 3a summarizes the results of χ^2 tests of heterogeneity in variances. For lathosterol there were no differences between sexes or generations in total variances, as indicated by the non-significant decreases in χ^2 in comparison to the reduction in the degrees of freedom (see Table 3a). For campesterol an almost significant sex difference (critical value for χ^2 with 1 *df* is 3.84) and for β-sitosterol a significant sex difference in variances was observed. Both for campesterol and for β-sitosterol a significant interaction of sex and generation was found (critical χ^2 value for 3 *df* is 5.76). For campesterol the χ^2 for a model that specified homogeneity in variances across sexes and generations was 47.58 and for the interaction model it was 40.48. The interaction model thus gives a significant improvement in likelihood when compared to the homogeneity model (χ^2 difference is 7.10 with 3 *df*). A similar improvement in likelihood was observed for β-sitosterol. The significant sex by generation interaction was due to a lower standard deviation in sons than in all other family members.

Correlations between family members are presented in Table 2. For all three variables, MZ correlations were consistently higher than the corresponding DZ and parent–offspring correlations, indicating that genetic factors are of importance. The correlations of DZ opposite-sex twins were not systematically lower than DZ same-sex correlations, indicating that a model in which the same genes and the same environmental factors influence lathosterol and phytosterol levels in males and females is appropriate (Heath et al., 1989).

In Table 3b χ^2 tests of differences in correlations between sexes and zygosities are summarized. For lathosterol a simple pattern of correlations emerged. There was no association between plasma lathosterol concentrations of fathers and mothers of twins as can be seen in Table 3b by the non-significant increase in χ^2 when spouse correlations were constrained to be zero (χ^2 difference with 1 *df* equals 37.50 – 36.80 = 0.70), which is a non-significant difference in likelihood). MZ and DZ correlations as well as parent-offspring did not depend on the sex of family members. However, the correlations for DZ twins were significantly larger than the parent-offspring correlations.

Results for campesterol indicated a significant correlation (r = .17) between spouses (χ^2 difference is 45.46 – 41.12 = 4.34 with 1 *df*). The MZ correlations were equal in males and females, as were the DZ correlations. Parent–offspring correlations also were independent of the sex of either the parent or the child. Parent–offspring correlations were not significantly different from DZ correlations.

The same pattern of familial correlations emerged for β-sitosterol as for campesterol, except that there was no significant association between β-sitosterol levels of spouses. However, as can be seen in Table 3, the χ^2s for all tests involving β-sitosterol levels were rather high and almost significant. This was mainly caused by heterogeneity between the 5 different family groupings in parent–offspring correlations.

Genetic model fitting was first carried out on the data of the twins without using the parental information. Table 4 lists the χ^2 and probability levels for different models of familial resemblance. These showed that for lathosterol an GCE model without sex differences, that is, a model with genetic, common and unique environmental sources of variation, gave a good account of the data (χ^2 = 10.85, p = .54). Leaving out either G or C did not lead to a significant decrease in fit, but specifying only E (i.e., a model of no resemblance between twins) lead to model that did not fit the data at all. However, it is not possible to distinguish on statistical grounds between a GE or CE model. In contrast, for both campesterol and β-sitosterol it is clear that a GE model gave the best account of the data. For these traits a scalar parameter was introduced to account for differences in total variances between sons and daughters, while constraining heritabilities to be the same in both sexes (Neale & Cardon, 1992). At the bottom of Table 4 the percentages of total variance explained by genetic factors, common environment and non-shared or unique environment are given. Heritability for lathosterol was 32% and the contribution of shared environment was of the same magnitude (34%). Heritabilities for the plant sterols were high, 83% for campesterol and 82% for β-sitosterol.

The results of genetic model fitting to data of parents and twins simultaneously are summarized in Table 5. For lathosterol a model with equal heritabilities in parents and offspring gave a heritability of 29%. The additional resemblance between twins for lathosterol was accounted for by shared environmental factors (37% of the total variance). This model gave a good fit to the data and showed a close

Table 1a

Means for Age (Years), Body weight (kg) and Untransformed Lathosterol, Campesterol and β-sitosterol (mmol/L) in Fathers, Mothers, Sons and Daughters

	N	Age	Weight	Lathosterol	Campesterol	β-sitosterol
Fathers	155	48.1	80.90	7.48	12.58	6.69
Mothers	156	45.6	68.00	6.11	12.32	6.95
Sons	158	16.8	61.54	3.87	11.83	6.36
Daughters	156	16.7	56.71	4.04	12.36	7.00

resemblance to the results obtained in the genetic analysis of the twin data only. However, a model with different heritabilities in parents and children without shared environmental influences gave an almost equally good account of the data (genetic heritability 10% in parents and 68% in children; (χ^2 = 47.32, *df* = 46, and *p* = .42). For campesterol and β-sitosterol a simple additive genetic model gave the most parsimonious account of the data. Resemblance between spouses for campesterol was attributed to the environmental component, but the fit of this model did not differ from a model in which phenotypic assortative mating was specified. Heritability estimates were 80% for campesterol and 73% for β-sitosterol and were almost the same as the estimates obtained from the analyses of the twin data only.

Table 1b

Standard Deviations for Log-transformed Lathosterol, Campesterol and *β*-sitosterol

	Lathosterol	Campesterol	β-sitosterol
Fathers	0.157	0.165	0.161
Mothers	0.161	0.173	0.169
Sons	0.153	0.141	0.135
Daughters	0.149	0.176	0.178

Table 2

Maximum-likelihood Estimates of Familial Correlations for Log-transformed Plasma Concentrations of Lathosterol, Campesterol and β-sitosterol

	N of Pairs	Lathosterol	Campesterol	β-sitosterol
MZM	35	.73	.89	.90
MZF	34	.60	.79	.76
DZM	30	.58	.50	.55
DZF	30	.50	.36	.25
DOS	28	.32	.44	.23
All MZ	69	.67	.84	.83
All DZ	88	.51	.46	.36
Spouses	155	.06	.17	.04
Father-son	155	.01	.26	.22
Father-daughter	157	.16	.24	.21
Mother-son	156	.22	.39	.38
Mother-daughter	157	.21	.27	.29
All Parent-child	155	.14	.30	.27

Note: MZM = Monozygotic Males, MZF = Monozygotic Females, DZM = Dizygotic Males, DZF = Dizygotic Females, DOS = Dizygotic Opposite-Sex Twins

Table 3

Heterogeneity Tests of Standard Deviations and Familial Correlations of Log-transformed Values for Lathosterol, Campesterol and β-sitosterol: χ^2 and Probability. Best Fitting Models Are Italicized.

A. Standard Deviations	Lathosterol			Campesterol		β-sitosterol	
	df	χ^2	*p*	χ^2	*p*	χ^2	*p*
No sex or generation differences	39	*36.80*	*(.57)*	47.58	(.16)	57.36	(.03)
Sex Differences	38	36.79	(.53)	43.79	(.24)	51.98	(.07)
Generation Differences	38	36.08	(.56)	46.60	(.16)	56.84	(.03)
Sex and generation Differences	36	35.87	(.48)	40.48	(.28)	47.82	(.09)
Separate SD for Sons	38	—	—	*41.12*	*(.34)*	*49.26*	*(.10)*
B. Correlations	**Lathosterol**			**Campesterol**		**β-sitosterol**	
	*df**	χ^2	*p*	χ^2	*p*	χ^2	*p*
Spouse correlation equal to zero	40	37.50	(.58)	45.46	(.22)	49.51	(.12)
Correlation MZM = MZF	41	40.48	(.50)	41.76	(.35)	52.30	(.09)
Correlation DZM = DZF	42	40.54	(.54)	42.59	(.36)	54.44	(.08)
Correlation DZM = DZF = DOS	43	40.71	(.57)	43.18	(.38)	57.04	(.06)
All parent-child correlations equal	46	*43.62*	*(.57)*	44.46	(.45)	59.92	(.07)
Correlation DZ = Parent-child	47	57.58	(.14)	*46.18*	*(.42)*	*60.33*	*(.08)*

Note: *degrees of freedom (*df*) given are for lathosterol; for β-sitosterol 1 *df* must be subtracted to account for lower variance in sons; for campesterol 2 *df* must be subtracted to account for lower variance in sons and for spouse correlation

. Values in italics indicate no variance difference for lathosterol and smaller variances in sons for campesterol and β-sitosterol. For lathosterol all parent–offspring correlations were independent of sex, for campesterol and β-sitosterol parent–offspring correlations equalled DZ correlations.

Discussion

We found significant familial aggregation for plasma levels of lathosterol and phytosterols in a sample of Dutch families consisting of twins aged 14–21 years and their parents aged 35–65 years. For lathosterol it proved difficult to distinguish between a model in which the observed familial resemblance was accounted for completely by shared genes with a higher heritability in children than in parents and a model in which heritabilities were the same in parents and children and in which the larger resemblance between siblings was accounted for by common environmental factors shared between them. If these results for lathosterol are compared to the results obtained by Berge et al. (2002) who found midparent–offspring correlations of 0.42, the model with equal heritabilities (29%) in twins and parents seems most plausible. There has been one other small twin study that examined the heritability of non-cholesterol sterol concentrations. Kesaniemi et al. (1989) studied a precursor of lathosterol in 17 MZ and 18 DZ Finnish twin pairs and found a heritability of around 50%.

In contrast to the Berge et al. study we observed additional resemblance in the twin offspring that could be attributed to shared environment. Both our study and the Berge et al. study observed high MZ correlations for lathosterol, but the DZ correlation (0.51) in our study was substantially higher than the DZ correlation (0.13) seen in Berge et al. study. Differences in sample size may play a role, but age differences could also be of importance. The Dutch twins were 17 years old on average and still lived at home with their parents, while the German twins who participated in the Berge et al. study had an average age of 25 years. The significant shared environmental component for lathosterol in adolescent twins might reflect the current shared family environment of twins. In addition, it might also reflect earlier shared influences or reflect the influence of a trait such as birthweight. We showed, both in this same sample of twins (Ijzerman et al., 2001) and in a much larger sample of young twins from the Netherlands Twin Register (Van Baal & Boomsma, 1998), that the influence of shared environment on birthweight is substantial. In a study of children who were born preterm Mortaz et al. (2001) reported that low birthweight was associated with

Table 4a

Model Fitting (χ^2 and Probability) to Log-transformed Values for Lathosterol, Campesterol and β-sitosterol in Twins

Model	Lathosterol			Campesterol		β-sitosterol	
	df*	χ^2	*p*	χ^2	*p*	χ^2	*p*
GCE:	12	*10.85*	*(.54)*	12.97	(.30)	20.90	(.03)
GE:	13	14.22	(.36)	*13.28*	*(.35)*	*20.90*	*(.05)*
CE:	13	13.81	(.39)	35.31	(.00)	48.77	(.00)
E:	14	72.51	(.00)	116.46	(.00)	117.62	(.00)

Note: **df* are given for lathosterol, for campesterol and β-sitosterol 1 *df* must be subtracted to account for sex differences in variances. G stands for additive genetic influences, C for common environmental factors shared by siblings and E for individual specific environment.

Best fitting models are italicized.

Table 4b

Model Fitting (χ^2 and Probability) to Log-transformed Values for Lathosterol, Campesterol and β-sitosterol in Twins

Variance components	Lathosterol	Campesterol	β-sitosterol
Heritability	32%	83%	82%
Shared environment	34%	—	—
Unique environment	34%	17%	18%

Table 5

Model Fitting to Log-transformed Lathosterol, Campesterol and β-sitosterol Data from Parents and Twins. Equal Heritabilities in Parents and Offspring. Additional Resemblance of Siblings for Lathosterol Is Explained by Environmental Factors Shared by Twins

	Lathosterol	Campesterol	β-sitosterol
χ^2	43.63	52.14	75.41
df	47	46	47
p	0.61	0.25	0.01
Variance Components			
Heritability	29%	80%	73%
Shared twin environment	37%	—	—
Environment	71%	20%	27%

an increased plasma lathosterol and a decreased campesterol at age 8–12 years. We tried to replicate these findings in our adolescent twin sample (Ijzerman et al., 2002), but did not find an association of birthweight with lathosterol or the phytosterols, neither in the overall sample, nor within twin pairs. The nature of the shared environmental factors which influence lathosterol levels in plasma thus remains to be identified.

For plasma plant sterol concentrations we found considerable heritability. Genetic influences accounted for 80% of the variance in plasma campesterol concentrations and for 73% of the variance in β-sitosterol levels. No influence of a common environment shared by family members was present for variation in phytosterol concentrations. Both campesterol and β-sitosterol reflect dietary cholesterol absorption. Plant sterols are not synthesized by humans and only a small percentage of plant sterols present in the diet is absorbed. Our results strongly suggest that sharing the same diet — as was to a large extent the case for the subjects in our study (Boomsma, 1990) — is not an important determinant of plasma phytosterol levels, although the high heritability estimate could also include genotype by shared environment interaction (Boomsma & Martin, 2002; Lynch & Walsh, 1998). If in the Dutch population phytosterol levels would be a valid reflection of fractional cholesterol absorption, the present study offers strong evidence for a genetic influence on cholesterol absorption. The present findings are in line with our previous report (Kempen et al., 1991) that subjects carrying the apo-E4 allele have higher phytosterol levels. Since dietary cholesterol may inhibit LDL-receptor synthesis (Sorci-Thomas et al., 1989), the LDL-receptor dependent clearance in the liver (Spady & Dietschy, 1985) and consequently increase plasma LDL-concentration (Appelbaum et al., 1984; Srivastava et al., 1991) we suggest this mechanism may be one of the possible causes for the high heritability of plasma LDL cholesterol (Boomsma et al., 1996) that we observed in the twins participating in this study.

Recently, two highly homologous genes located on chromosome 2p21 have been found to be involved in sitosterolemia (Berge et al., 2000; Lu et al., 2001). Patients with sitosterolemia have very elevated levels (~ 50-fold elevations) of plant sterols and are at risk for hypercholesterolemia and premature atherosclerosis (Wittenburg & Carey, 2002). The two genes encode ATP binding cassette (ABC) half transporters ABCG5 and ABCG8. Sitosterolemia patients have mutations in one of these genes. These mutations are rare in the general population, but it may be that more common, milder, variants in these genes explain part of the significant heritability seen for phytosterol concentrations. Berge et al. (2002) found in their general population sample that two sequence variants in ABCG8 were associated with plant sterol levels. The D19H polymorphism in exon 1 was associated with campesterol-cholesterol ratios and the T400K polymorphism was associated with sitosterol-cholesterol ratios. For both polymorphisms the rare allele was associated with lower sterol levels. The amount of variance explained by these polymorphisms, however, did not explain all of the heritability in plant sterol concentrations. Thus, polymorphisms at other loci besides ABCG5 and ABCG8 probably contribute to variation in plant sterol levels in the general population. These polymorphisms could be detected in a systematic scan of the entire genome. We are currently in our group of DZ twins collecting marker data (Beekman et al., 2003) for linkage analysis. However, the power of linkage analysis in small, unselected, samples is low. Genetic association analysis of candidate genes identified in animal studies might be a more powerful alternative. The ABCG5 and ABCG8 genes which map to mouse chromosome 17 have both found to influence the fractional absorption of dietary plant sterols in mice (Yu et al., 2002a, 2002b). However, a linkage study of campesterol and sitosterol levels in mice identified two other loci on mouse chromosome 2 and a major locus on mouse chromosome 14 (Sehayek et al., 2002), which could be candidates to explain the heritability of plant sterol levels in humans.

For lathosterol levels, which seem to be characterized by a lower heritability than phytosterol concentrations, no candidate gene studies have yet been published. However, a recent publication (Brunetti-Pierri et al., 2002) describes in a lathosterolis patient two mutations in the SC5D gene, which is involved in the conversion of lathosterol into 7-dehydrocholesterol. The activity of 3β-hydroxysteroid-Δ5-desaturase (SC5D), the enzyme involved in this reaction, was deficient in the patient's fibroblasts. Sequence analysis of the SC5D gene in the patient's DNA showed two missense mutations, indicating that the patient is a compound heterozygote. The mutations were not present in 50 unaffected individuals, but the gene may harbour milder variants that are involved in lathosterol variation in the general population.

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