5. DETERMINANTS OF PLASMA HOMOCYSTEINE

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SUMMARY

The concentration of total homocysteine in plasma is influenced by a diversity of genetic and acquired factors, and by interactions between such factors. The most prevalent genetic cause of hyperhomocysteinemia is the C677T polymorphism of the methylenetetrahydrofolate reductase gene, which predisposes to hyperhomocysteinemia under conditions of impaired folate status. Among the physiological and life-style determinants, increasing age, male sex, poor nutrition with low vitamin intake, smoking, heavy coffee consumption cause high homocysteine, whereas young age, premenopausal state, pregnancy, vitamins like folate and cobalamin, and exercise are associated with low homocysteine. Several drugs may influence the homocysteine level by acting as vitamin antagonists, and among these, methotrexate and nitrous oxide cause a rapid increase in homocysteine by interfering with folate and cobalamin functions, respectively. Some sulfhydryl-containing drugs reduce homocysteine, probably via disulphide exchange reactions, whereas the effect of steroid hormones on homocysteine is complex and their mechanisms are conjectural. Cyclosporin A increases homocysteine, possibly by a mechanism independent of interference with renal function. The diseases which most often and profoundly increase homocysteine are folate and cobalamin deficiencies and renal failure. Some proliferative (psoriasis) and malignant (leukemia) diseases may increase homocysteine, probably by directing folates towards DNA synthesis. Hyperhomocysteinemia has been

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associated with diabetes, but this is most likely secondary to impaired renal function, since factors like insulin itself and glomerular hyperfiltration seem to reduce homocysteine.

Table 5-1. Categorization of plasma homocysteine determinants

Categories
Genetic factors
Physiological determinants
Life-style factors
Diseases
Drugs

Other states that affect homocysteine are thyroid dysfunction, heart transplantation and the acute phase after a cardiovascular event. The implications of variations of homocysteine according to various determinants are threefold. First, elevated homocysteine may be useful for diagnosis or follow up of some diseases or drug therapies, as for example folate or cobalamin deficiencies or nitrous oxide anaesthesia. Secondly, high homocysteine may itself be hazardous by predisposing to occlusive vascular disease, and may contribute to the increased prevalence of cardiovascular disease in conditions like renal failure or hypothyroidism. Finally, strategies to modify factors predisposing to hyperhomocysteinemia may have a health promoting effect, and are actually in line with established guidelines promoting good health.

INTRODUCTION

Plasma total homocysteine levels are determined by a variety of factors. These factors are categorized in table 1. This chapter will focus on physiological and life-style determinants, effect of drugs and various diseases, with emphasis on the Hordaland Homocysteine study, a population based screening of 18043 healthy subjects aged 40-67 years. The relation of plasma homocysteine to genetic factors, vitamin status and renal function will be addressed in detail in Chapters 6 and 15-18 are only briefly summarized here.

GENETICS

Heterozygosity for cystathionine ß-synthase (CBS) deficiency was established as a cause of elevated post-methionine loading plasma homocysteine more than 30 years ago (1). The fasting levels in these individuals seem to be normal or slightly elevated (2). It has been suggested that heterozygosity for CBS deficiency explains hyperhomocysteinemia in most vascular patients (3, 4), but this conclusion has later been refuted (5). Although post loading homocysteine seems to be partly a genetic trait (6), the frequency of carriers of CBS deficiency is not sufficiently high to account for hyperhomocysteinemia in either a normal (7) or vascular population (8).

Inheritance as a factor influencing fasting plasma homocysteine was suggested by the finding of a correlation between the homocysteine levels in monozygotic and dizygotic healthy twins (9, 10), and between family members with heart disease (6, 11, 12). Healthy children (13) and children with familiar hypercholesterolemia (14) who have relatives with cardiovascular disease, have higher fasting homocysteine than children with healthy relatives, suggesting that the fasting level is a genetic transmittable risk factor.

The common C677T polymorphism of the methylenetetrahydrofolate reductase (MTHFR) gene has been established as an important genetic determinant of elevated fasting homocysteine. Homozygosity for this polymorphism (TT genotype) predisposes to hyperhomocysteinemia under conditions of impaired folate status (Chapter 16). Also in children, homocysteine is determined by the MTHFR status, and the highest levels are observed in TT subjects with low serum folate (15). Mutations in the methionine synthase gene causing moderate hyperhomocysteinemia have hitherto not been identified (16).

PHYSIOLOGICAL DETERMINANTS (FIG. 5-1).

Plasma homocysteine increases throughout life in both sexes. Before puberty, homocysteine in children is similar in boys and girls (about 5 µmol/L). The levels show a marked increase, particularly in boys, between pubertal stages 1 and 2-5. By puberty, the plasma homocysteine level has increased to about 6-7 µmol/L (13-15, 17-19), and the characteristic skew distribution (13) and the sex difference observed in adults have been established.

In adults, the plasma homocysteine levels are usually about 1-2 µmol/L higher in men than in women. In the Norwegian Hordaland cohort, the geometric means were 10.8 µmol/L in 5918 healthy men and 9.1 µmol/L in 6348 women aged 40-42 years (7). From puberty to old age, mean homocysteine increases (about 3-5 µmol/L) in both sexes (7, 20), but homocysteine seems to

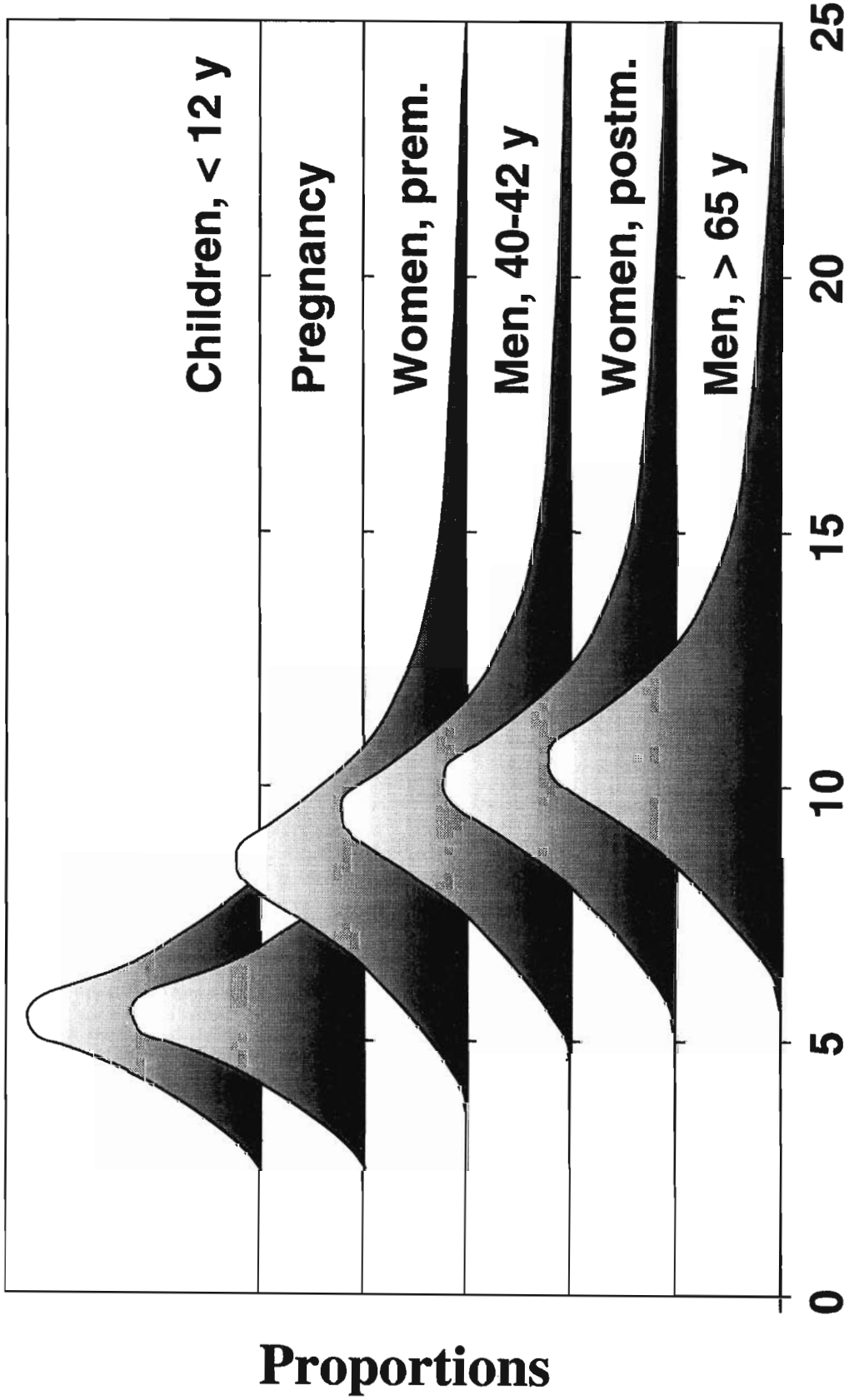

Figure 5-1 Plasma homocysteine level and distribution according to physiological factors.

decline in the very old (21). The age-related increase in homocysteine is steeper in women than in men. This is at least partly seems to be related to menopause. Both fasting and post methionine load homocysteine are higher in postmenopausal than in premenopausal women (20, 22). The age-dependent increase may be attributed to deterioration of renal function (23, 24) and impaired folate status (25, 26), whereas the sex-related differences are explained by the effects of sex steroids on homocysteine or possibly higher homocysteine production (linked to creatine-creatinine synthesis (2)) in men due to higher muscle mass (see later).

There are consistent reports of a substantial reduction (by about 50%) of homocysteine during pregnancy (27, 28). The reduction seems independent of folate status (29). homocysteine decreases between the first and second trimester, and thereafter remains essentially stable throughout the rest of the pregnancy (28). After delivery, the maternal homocysteine level is inversely related to neonatal weight and gestational age (30). Normal homocysteine concentrations are attained 2-4 days post-partum (28). An umbilical vein to artery homocysteine decrement and a relation between homocysteine and neonatal weight and gestational age have been interpreted as fetal uptake of maternal homocysteine (30). Alternatively, low homocysteine may represent physiological adaption to pregnancy (31), which may support adequate placental circulation.

LIFE-STYLE AND DIET (FIG. 5-2)

The metabolic relations between homocysteine and methionine (Chapter 3) and the metabolic adaption to methionine excess in the rats (32), raise the possibility that methionine intake may influence fasting or post methionine load homocysteine. Plasma homocysteine increases by about 14% 8 hours after a protein rich meal (33). However, neither the homocysteine response after loading (34, 35) nor fasting homocysteine (36) seems to be related to the daily dietary methionine or protein content. On the contrary, there are two reports suggesting that high dietary protein intake (37) or methionine intake (38) may actually decrease fasting homocysteine. This observation is in accordance with infrequent elevation of homocysteine and positive cobalamin status in high meat-eaters (39).

Folate and cobalamin deficiencies are common causes of moderate to severe fasting hyperhomocysteinemia (40, 41), whereas vitamin B_6 deficiency normally results in increased post methionine load homocysteine only (42). Fasting plasma homocysteine correlated negatively with both intake and serum levels of folate, cobalamin and vitamin B_6 (36, 43). These relations between vitamin status and homocysteine are corroborated by supplementation studies

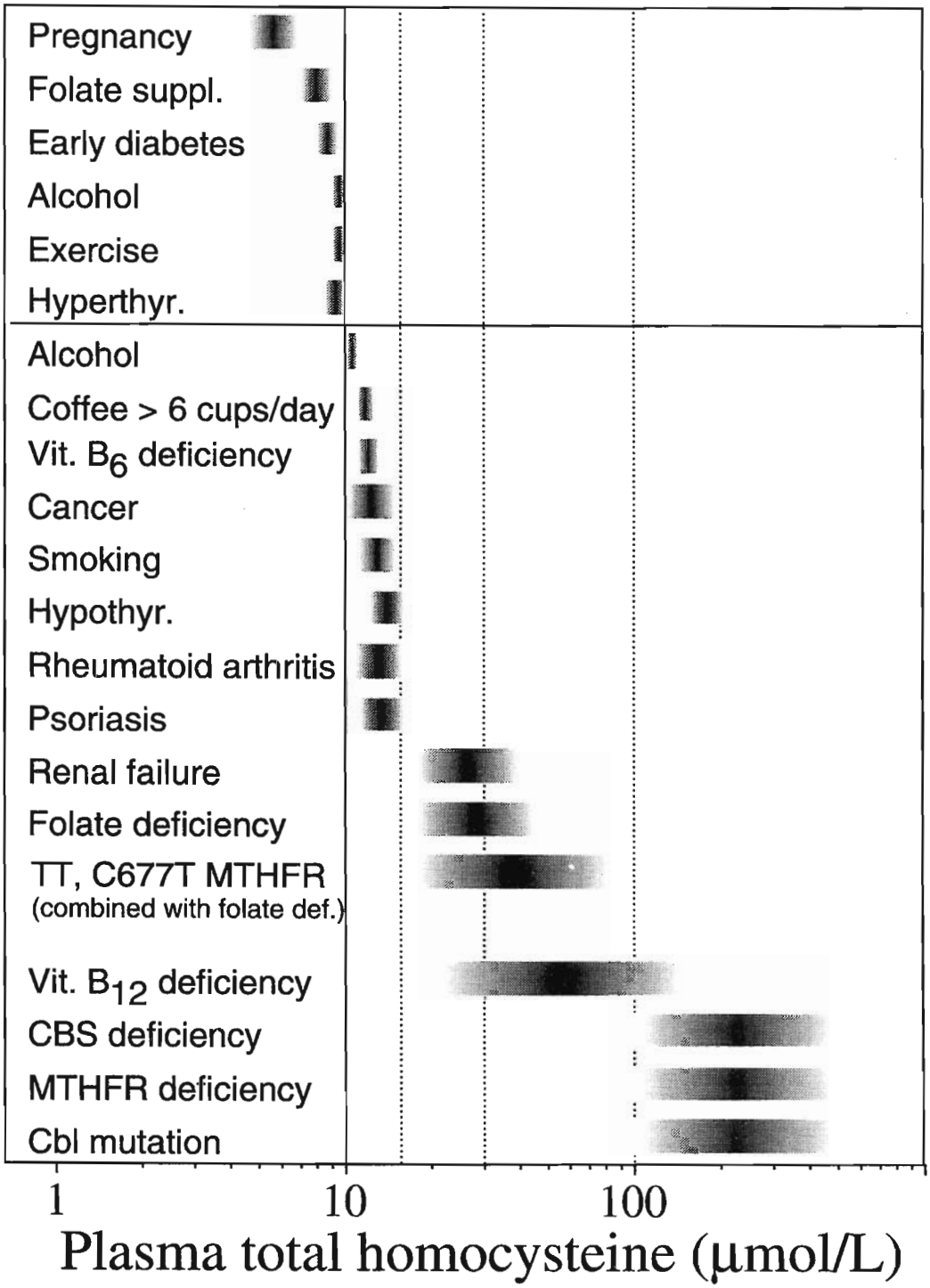

Figure 5-2. Schematic representation of the effect from an isolated determinant on the expected mean homocysteine level. Normal value for homocysteine is defined as 10 µmol/L, and factors that reduce and increase homocysteine are sorted and separated by the horizontal line. The width of the bars does not indicate the range of homocysteine values but rather the uncertainty of the estimate, related to the extent or severity of disease or variable response. The estimates are not based on published data, but rather reflect an overall impression of the authors. Relevant literature is quoted in the text.

showing that folate is the most efficient means to reduce fasting homocysteine (41, 44), whereas vitamin B_6 does not affect fasting homocysteine (45) but selectively influences post load homocysteine (41).

Folic acid supplementation seems to be more efficient in lowering homocysteine than folate derived from food (46, 47), and a meta-analysis of intervention studies demonstrated that increasing the folic acid dose above 0.5 mg/d does not further reduce the homocysteine concentration (44). The effectiveness of folate supplementation seems to reach a plateau at about 0.4 mg/day (47), and 0.2-0.4 mg/day have been reported to maintain a positive folate homeostasis and thereby optimal homocysteine remethylation in healthy subjects (48, 49). A similar dose-response relationship has recently been observed for folic acid fortified cereals demonstrating maximal homocysteine lowering effect between 0.5 and 0.665 mg folic per 30 gram cereal (50).

The homocysteine distribution in the general population is skew with a marked tail towards higher homocysteine values (7). There are consistent reports that homocysteine is reduced and approaches normal distribution both after folic acid supplementation and in subgroups with adequate vitamin status (51-53). In the Hordaland Homocysteine cohort, we could distinguish between the reduction of high homocysteine to normal levels, which is usually conferred by folate derived from food, and the reduction from normal to subnormal levels which is attributable to intake of folic acid containing supplements (54).

In addition to the expected effect of dietary folate, recent studies have also provided data which demonstrate that life-style significantly affects the plasma homocysteine level (54).

Higher levels of homocysteine in smokers than in non-smokers have been demonstrated in some (12, 55, 56) but not all (8) smaller studies. The Hordaland Homocysteine study demonstrated a strong dose-response relationship between the number of cigarettes and homocysteine levels, independent of age and sex (7), also in subjects with high folate intake (54). Notably, smoking increases mean homocysteine and causes a shift of the whole homocysteine distribution curve to higher levels, similar to that observed in populations with low folate intake (54). This may suggest an influence of smoking on folate function. However, smokers generally consume a less healthy diet containing less vegetables and more fat than non-smokers (57-59), and smokers have reduced intake and blood levels of several vitamins involved in homocysteine metabolism, including vitamins B_{12} (60) and B_6 (61, 62). In addition, tobacco smoke contains abundant free radicals that confer oxidative stress and thereby may affect redox status of thiols (63), including homocysteine (64).

Heavy coffee consumption was among the strongest life-style determinants of homocysteine in the Hordaland Homocysteine cohort (65). A dose-response relation was observed, and in individuals drinking more than 6 cups each day, the mean homocysteine level was 2-3 μmol/L higher than in coffee abstainers.

In contrast, in US participants in the ARIC study, there was no relation between homocysteine and moderate coffee consumption. A recent study demonstrated homocysteine elevation in the elderly consuming 4 or more cups daily (37).

Coffee consumption is known to be associated with unhealthy life-style and poor nutrition (57, 66, 67), but the homocysteine-coffee relation reported in the Hordaland Study was also found in non-smokers and at both high and low folate intake. Heavy coffee consumption increases mean homocysteine by decreasing the proportion with low and intermediate homocysteine, and in this respect can be distinguished from folate deficiency and cigarette smoking (54). Notably, the effect was observed with filtered coffee and thus is not mediated by the cholesterol-raising diterpenes. As the consumption of decaffeinated coffee did not have an effect on homocysteine, caffeine may play a mechanistic role (65). The caffeine effect may be related to its influence on the cardiovascular system or kidney function (68). Another possibility is interference with vitamin B_6 function, as reported for another xanthine, theophylline (69), but such a mechanism implies a predominant effect on post methionine load homocysteine.

In the Hordaland study, a life-style profile, which reflects the combined effect of the three major modifiable homocysteine determinants, folate intake, smoking and coffee consumption, is strongly correlated with homocysteine (54). Subjects with a contrasting life-style have a difference of 3-5 μmol/L in homocysteine which is larger than the effect attributable to each factor alone. This supports the notion of different mechanisms underlying the homocysteine elevating effects of smoking, low folate intake and heavy coffee consumption. Furthermore, homocysteine is essentially normally distributed in a population characterized by a healthy life-style profile (54).

Among the 18043 subjects investigated in the Hordaland Homocysteine study, only 67 (0.4%) had homocysteine equal or higher than 40 μmol/L (70). Compared to controls, these subjects had lower plasma folate and cobalamin levels, lower intake of vitamin supplements, consumed much coffee and were frequently (60%) smokers. When 7 subjects with cobalamin deficiency were excluded, 92% of these hyperhomocysteinemic subjects (compared to 10.4% controls) were homozygous for the C677T MTHFR polymorphism. These findings demonstrate a strong positive interaction between MTHFR genotype and life-style determinants of homocysteine (70).

Both exercise and moderate alcohol consumption are weak but significant determinants of homocysteine in the Hordaland cohort (7, 71). The difference in homocysteine between subjects with sedentary life-style compared with those doing exercise on a daily basis is most pronounced in the elderly, and approaches 1 μmol/L. Exercise reduces the skewness of the homocysteine distribution curve, and therefore seems to lower homocysteine in subjects with

Table 5-2. Drug effects on plasma total homocysteine

Class Drug	homocysteine response	Possible mechanism
Folate antagonists		
Methotrexate	Increase	Inhibition of DHFR[a], depletion of reduced folates
Anticonvulsants	Increase	Inhibition of polyglutamation, folate depletion
Colestipol	Increase	Inhibition of folate absorption
Cholestyramine	Increase	Inhibition of folate absorption
Cobalamin antagonists		
Nitrous oxide	Increase	Inactivation of methionine synthase
Nitric oxide	ND[a]	Inactivation of methionine synthase
Metformin	Increase	Inhibition of cobalamin absorption
H2-receptor antag.	ND	Inhibition of cobalamin absorption
Omeprazole	ND	Inhibition of cobalamin absorption
Cholestyramine	ND	Inhibition of cobalamin absorption
Vitamin B_6 antagonists		
Niacin	Increase	Inhibition of pyridoxal kinase
Azauridine	Increase	Inhibition of pyridoxal kinase
Isoniazid	ND	Inhibition of pyridoxal kinase
Theophylline	Increase	Inhibition of pyridoxal kinase
Homocysteine production		
Adenosine analogues	Decrease	Inhibition AdoHcy[a] hydrolase
L-Dopa	Increase	Substrate for AdoMet[a]-dependent transmethylation
Sulfhydryl compounds		
D-Penicillamine	Decrease	Disulphide exchange, displacement
N-Acetylcysteine	Decrease	Disulphide exchange, displacement
Mesna	Decrease	Disulphide exchange, displacement
Sex steroids		
Contraceptives	Increase, variable	Not known, interference with vitamin
Estrogens (postm.)	Decrease	Not known, interference with vitamin function
Androgens	Increase	Increased muscle mass and creatinine synthesis
Antiestrogens		
Tamoxifen	Decrease	Not known
Aminoglutethimide	Increase	Induction of liver metabolism
Others		
Cyclosporin A	Increase	Impaired renal function
Betaine	Decrease	Enhancement of remethylation

[a]Abbreviations: ND, not determined; DHFR, dihydrofolate reductase; AdoHcy, S-adenosylhomocysteine; AdoMet, S-adenosylmethionine

hyperhomocysteinemia (7). In subjects aged 40-42 years, and in particular among smokers, the relation between homocysteine levels and long-term

alcohol consumption forms a weak U-shaped curve with reduction in homocysteine up to 14 alcohol units per week. Higher alcohol intake increases homocysteine (71).

Plasma homocysteine shows a transient increase during acute alcohol intoxication in alcoholics (72), and direct inhibition of methionine synthase by acetaldehyde (73) should be considered as a possible mechanism. Chronic alcoholism seems to be associated with hyperhomocysteinemia (74), but only in subjects with poor nutrition (72). This may be explained by impaired folate, vitamin B_{12} or vitamin B_6 status (74).

DRUGS (TABLE 5-2)

A variety of drugs affect homocysteine levels. They act via different mechanisms, including inhibition of vitamin (folate, cobalamin or vitamin B_6) function, by affecting homocysteine production, undergoing thiol-disulphide exchange reactions, interfering with renal function and influencing hormonal status. Most literature on homocysteine and drugs has been reviewed previously (75-77). The effects of various drugs on homocysteine are summarized in Table 5-2.

The antifolate drug methotrexate (MTX) induces elevated homocysteine within hours after high-dose infusions (up to 33.6 grams /m^2) used for cancer chemotherapy, and this effect is subsequently reversed by rescue therapy with high doses of folinic acid (78, 79). In leukemia patients, the marked increase in plasma homocysteine was associated a 4-fold increase in cerebrospinal fluid homocysteine and a massive build-up (from undetectable levels to about 30-100 μmol/L) of homocysteic and cysteic acid in the cerebrospinal fluid. Accumulation of these neuroexcitatory amino acids is related to and may be responsible for neurological toxicity (80).

In psoriasis patients treated with low doses of methotrexate (10-25 mg), homocysteine increases slowly over a period of several days (81). In rheumatoid arthritis patients treated with 1 mg/day methotrexate, elevated homocysteine seems to develop slowly between 3-6 months (82) and one year (83). Folic acid (5-27.6 mg/week) improves folate status and prevents the methotrexate-induced hyperhomocysteinemia (83) and toxicity, but preserves the therapeutic efficacy in rheumatoid arthritis patients (84).

Several other drugs may cause hyperhomocysteinemia through interference with folate metabolism. This has been demonstrated for phenytoin and other anticonvulsants (75, 85, 86), which probably act by depleting liver folate stores by inhibiting polyglutamation (87). Plasma homocysteine is also elevated following therapy with niacin in combination with the bile sequestrant colestipol.

The latter agent may interfere with folate absorption (88). Notably, elevation of homocysteine following treatment with another bile resin, cholestyramine, was largely confined to subjects with the C677T transition in the MTHFR gene (89). This genotype predisposes to hyperhomocysteinemia under conditions of impaired folate status.

Plasma homocysteine increases within hours in patients exposed to the anaesthetic gas, nitrous oxide (90-94). The increase reflects irreversible oxidation of cob(I)alamin formed as a transient intermediate of the methionine synthase reaction. This, in addition to the irreversible inactivation of the enzyme methionine synthase itself, is assumed to be responsible for the side effects from bone marrow and central nervous system observed after prolonged nitrous oxide exposure (95). The deleterious effect of nitrous oxide on methionine synthase may be alleviated by methionine loading prior to anaesthesia (91).

The endothelial-derived relaxing factor, nitric oxide, has a similar effect on isolated methionine synthase (96, 97) and on the enzyme in isolated hepatocytes (98), but the significance of this interaction in vivo remains to be established.

In contrast to the rapid increase in homocysteine observed during nitrous oxide exposure, a slow increase over months to years is expected during prolonged intake of drugs interfering with cobalamin absorption. Such interference with cobalamin absorption has been reported for cholestyramine (99), histamine H2-receptor antagonists (100), omeprazole (101) and the antidiabetic metformin, but elevation of homocysteine has hitherto only been reported for cholestyramine (89) and metformin (102, 103). The latter two drugs may also affect folate absorption.

Several drugs interfere with the function of vitamin B_6. A common mechanism involves inhibition of pyridoxal kinase (104). High plasma or urinary homocysteine or homocystine has been reported following treatment with azauridine (105), isoniazid (106), niacin (107) and theophylline (69).

Some drugs may influence homocysteine production. Several adenosine analogues with antiviral properties inhibit S-adenosylhomocysteine hydrolase (108) and may thereby decrease homocysteine production. Such drugs are not widely used, and low plasma homocysteine has only been demonstrated for the antimetabolite, 2-deoxycoformycin (109). Enhanced homocysteine production may occur following intake of drugs that serve as a substrate for S-adenosylmethionine-dependent transmethylation reaction, as demonstrated for the antiparkinson drug, L-dopa, in rats (110) and humans (Miller and Brattström, unpublished). One may speculate whether a similar mechanism contributes to the hyperhomocysteinemia induced by niacin (107).

Three sulfhydryl-containing drugs, D-penicillamine (111, 112), N-acetylcysteine (113) and 2-mercaptoethane sulfonate (mesna) (114, 115), have been found to reduce plasma homocysteine. These drugs probably act by a

thiol-disulphide exchange reaction, which may enhance excretion, lower plasma protein-binding or alter distribution of homocysteine (77).

The effect of sex steroid hormones on homocysteine is indicated by sex differences in homocysteine level and by the observation of low homocysteine levels in premenopausal women (20, 116) and during pregnancy (27, 28). Inconsistent data (117-119) have been published on change in plasma homocysteine of women taking oral contraceptives (117), the effect of which seems to depend on the hormonal phase (118). Replacement therapy containing estrogen in postmenopausal women results in a decrease in plasma homocysteine within 3-6 months of treatment (120), after which the homocysteine returns to base-line in some (121) but not all patients (122). The mean decrease in homocysteine was 13.5%, but the largest reduction was obtained in women with highest base-line values (123).

Estrogen treatment reduces homocysteine of healthy men (124) and men with prostatic carcinoma (117), whereas short-term treatment of normal men with supraphysiological doses of testosterone is without effect (125). A recent cross-sex hormone study in transsexual males and females demonstrates that plasma homocysteine decreases after estrogen plus antiandrogen administration to male subjects, and increases after androgen administration to female subjects (126). This study suggests that physiological levels of sex hormones affect plasma homocysteine concentration. A positive correlation between homocysteine and plasma creatinine levels during androgen administration (126) suggests that androgens act by enhancing synthesis of creatinine and thereby homocysteine secondary to increase in muscle mass (126). In addition, sex hormones and contraceptives may impair folate (127), cobalamin (128) and vitamin B_6 status (129), which may predispose to hyperhomocysteinemia.

In postmenopausal breast cancer patients, the antiestrogens tamoxifen and aminoglutethimide have opposite effects on the homocysteine levels. Tamoxifen lowers plasma homocysteine after 6-12 months of treatment, particularly in subjects with high pre-treatment levels (130, 131). The drug also possesses estrogen agonistic effect, and the mechanism behind the homocysteine reduction is uncertain (130). Among three aromatase inhibitors which block the androgen to estradiol conversion, only aminoglutethimide causes a substantial increase in homocysteine. Thus, this effect is probably not related to low estrogen levels, but may be due to the ability of aminoglutethimide to induce hepatic mixed function oxidase (132), which has been associated with enhanced folate turnover (133).

The immunosuppressive drug cyclosporine A (CyA) increases plasma homocysteine. Renal transplant patients receiving CyA have significantly higher homocysteine than both non-treated renal transplant recipient, and patients without a renal graft but matched for glomerular filtration rate (134). In renal patients, it may be difficult to distinguish high homocysteine caused by CyA

interference with renal function from high homocysteine related to impaired renal function from other causes (135). Hyperhomocysteinemia also develops in cardiac transplant patients, and high homocysteine is predicted by both serum creatinine and serum CyA concentration (136), suggesting that the CyA effect is independent of renal function. The usual correlation between homocysteine and serum folate was absent in the CyA-treated renal transplant recipients, which may suggest a mechanism involving interference with folate-dependent remethylation (134). This conclusion has been refuted by a recent study (135), and is not supported by the observation of a reduction of homocysteine in CyA-treated renal transplant recipients by high-dose folic acid (137).

Betaine is the co-substrate in the betaine-homocysteine methyltransferase reaction (32), and has been extensively used as a safe and effective homocysteine lowering agent in homocystinuria. In contrast to vitamin B_6, folate or cobalamin, betaine is effective in all forms of homocystinuria (2). Data on betaine treatment of moderate hyperhomocysteinemia are sparse. Betaine in doses up to 6 g/day has been shown to reduce (138) or normalize (139, 140) post load homocysteine in vascular patients, but has essentially no effect on fasting homocysteine level in renal patients receiving folic acid (141). Betaine as possible means to reduce homocysteine should be further investigated.

DISEASES (FIG. 5-2)

Folate and cobalamin deficiencies and renal failure (142, 143) are the clinical states most often responsible for markedly elevated homocysteine. These conditions are discussed in detail in chapters 6 and 14.

High homocysteine has been demonstrated in children with acute lymphoblastic leukemia (79) and in patients with psoriasis (81). These are conditions with a large burden of proliferating cells which export more Hcy than resting cells (144), possibly due to drainage of the folate pool in the direction of DNA synthesis at the expense of homocysteine remethylation.

Data on homocysteine levels in rheumatoid patients are somewhat controversial, probably because of frequent systemic manifestations combined with variable and extensive drug treatment. In patients not receiving methotrexate, one small study has reported an elevated post load homocysteine level, attributable to impaired vitamin B_6 status often seen in rheumatoid arthritis (145). Normal fasting homocysteine has been found in patients not receiving long-term methotrexate (82, 145), whereas elevated fasting levels have been reporting in patients with severe and long-standing rheumatoid arthritis associated with impaired cobalamin absorption and function (146).

In type I diabetes, hyperhomocysteinemia occurs at an advanced stage and is characterized by elevated creatinine or macroalbuminuria. Elevated homocysteine is attributable to impaired renal function (147-149), but marginal folate deficiency may also contribute (150). In both type 1 and 2 diabetes, elevated fasting (151-153) or post methionine load homocysteine (152, 154) are associated with macroangiopathy, whereas a relation between homocysteine and microangiopathy (152, 155, 156) or microalbuminuria (157, 158) has been demonstrated in some but not all (148) studies. In type I diabetes patients with normal creatinine (159) and in non-diabetic hyperinsulinemic subjects (160), subnormal homocysteine has been reported. Low homocysteine may be due to the glomerular hyperfiltration observed in early diabetes (173) or is possibly a metabolic effect of high insulin level. The latter possibility is in agreement with elevated homocysteine in insulin-resistant subjects (161) and with reduction of homocysteine by insulin, as demonstrated during hyperinsulinemic-euglycemic clamp. The homocysteine reduction was not observed in type 2 diabetes, suggesting impaired insulin effects on homocysteine in these patients (162).

Homocysteine has recently been reported to be moderately elevated in hypothyroidism and low in hyperthyroidism (163). This may be related to the influence of thyroid status on riboflavin (164) or folate function, GFR or creatinine synthesis.

There are consistent reports on higher homocysteine in heart transplant recipients than in controls (165, 166), and close to 70% of these patients have values higher than the 90th percentile of controls. The increase in homocysteine takes place in the early postoperative phase and persists thereafter, and may be partly related to impaired functions of folate, B_6 status (167) or possibly vitamin B_{12} (166). Elevated homocysteine was related to vascular complications in one (168) but not all (167, 169) studies, and underlying low B_6 may be an independent predictor of cardiovascular morbidity and mortality (169).

Plasma homocysteine is low in the acute phase (first days) after myocardial infarction (38, 170, 171) or stroke (172) compared to the convalescent stage. In patients with infarction, homocysteine increases by about 40% within 7 days, and thereafter is essentially stable for at least 6 months or decreases slightly (171). The low homocysteine level in the acute phase is probably a response to the acute stress causing both hemodynamic and hormonal changes, but the possibility that the low homocysteine reflects the pre-infarction level cannot be ruled out.

Table 5-3. Common causes of various degrees of hyperhomocysteinemia.

homocysteine level	**Prevalence**[a]	**Common cause**[a]
Moderate elevation (15-30 µmol/L)	≤10%	Unhealthy life-style, including poor nutrition MTHFR[b] polymorphism combined with low folate status (S-folate in lower normal range) Folate deficiency Mild cobalamin deficiency Renal failure Hyperproliferative disorders Drug effects
Intermediate elevation (30-100 µmol/L)	≤1%	MTHFR polymorphism combined with folate deficiency Moderate cobalamin deficiency Severe folate deficiency Severe renal failure
Severe elevation (>100 µmol/L)	≤0.02%	Severe cobalamin deficiency CBS[b]-deficiency (homozygous)

[a]Prevelance data (for a normal population) taken from ref. 7. The prevalence and causes of high homocysteine may vary according to population investigated.

[b]Abbreviations: MTHFR, 5,10-methylenetetrahydrofolate reductase; CBS, cystathionine-ß-synthase.

SUMMARY AND CONCLUSION

Plasma homocysteine levels are related to physiological parameters like age, gender, and altered hormonal status during pregnancy and after menopause (Fig. 5-1). Knowledge of such variations forms the basis for the assessment of homocysteine status and for establishing reference intervals. Moderate changes of homocysteine concentrations of 1-4 µmol/L are secondary to several modifiable life-style factors, such as smoking, nutrition, vitamin intake, coffee consumption and exercise, and reduction of homocysteine should be an incentive to improve life-style. Such recommendations should be given irrespective of homocysteine being a cause or indicator of cardiovascular or other diseases, since they conform with established guidelines promoting good health. Some diseases, like renal failure and hypothyroidism, are associated with hyperhomocysteinemia, which may contribute to the increased cardiovascular morbidity in these patients. Several drugs elevate homocysteine. For methotrexate and nitrous oxide, homocysteine may be a valuable indicator of pharmacodynamics, and high homocysteine induced by

some drugs may confer increased cardiovascular risk (77). Folate and cobalamin deficiencies and some inborn errors of homocysteine metabolism cause a sub- stantial elevation of homocysteine, which serves as a disease indicator useful for diagnosis and follow-up. Finally, knowledge of the expected mean homocysteine level in the presence of various determinants (Fig. 5-2) and the prevalence of the determinants forms the basis of the diagnostic value of elevated homocysteine (Table 5-3).

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