Atherosclerosis, serum cholesterol and the homocysteine theory: a study of 194 consecutive autopsies.

Editor's Comment:
Dr. McCully must be lauded for his pioneering studies that led him to propose in 1969 what has become to be known as the "homocysteine theory of atherosclerosis". Knowing back then that the medical community was fixed on cholesterol as the leading cause of atherosclerosis Dr. McCully set out to prove his theory. If cholesterol were responsible for arterial narrowing then everyone with atherosclerosis would have high cholesterol levels. That this was wrong required Dr. McCully to initiate the study that follows. He found that most patients dying from severe atherosclerosis had cholesterol levels within the normal range. This further opened the door for alternative explanations and Dr. McCully had one, homocysteine.

Dr. McCully had already studied the severe atherosclerosis that occurs in patients with a metabolic disorder leading to extremely high blood levels of homocysteine, homocysteinuria. These patients inherited a gene each from mother and father and had a "double dose of the gene" or homozygous inheritance. He theorized that there were unrecognized "single dose" or heterozygous individuals walking around with elevated levels of homocysteine, those inheriting only one gene for high homocysteine, not knowing that they were at risk for developing atherosclerosis, heart attack, stroke, and blood clots.

A retrospective study examined 194 consecutive autopsies to determine the proportion of cases of atherosclerosis without elevated serum cholesterol, diabetes mellitus, or hypertension. The study cases were classified into four groups, according to the cause of death and the degree of atherosclerosis. Cases in Group 1, in which death resulted from complications of severe atherosclerosis, have a mean serum cholesterol of 186.7 +/- 41.8 mg/dL, and the cholesterol is less than 200 in 65% and less than 250 in 92% of cases. Cases in Group 2, with severe atherosclerosis dying of other diseases, have a mean serum cholesterol of 174.6 +/- 60.4 mg/dL, and the cholesterol is less than 200 in 79% of cases and less than 250 in 89% of cases. Cases in Groups 3 and 4, with moderate and minimal atherosclerosis, respectively, have mean serum cholesterol values of 172.3 +/- 54.8 and 143.5 +/- 47.8 mg/dL, and the cholesterol is less than 200 in 71% and 92% and less than 250 in 92% and 96% of cases, respectively. Serum cholesterol is significantly associated with severity of atherosclerosis in the total sample (P = 0.01). Three fourths of all cases (147/194) have neither diabetes nor hypertension, and in 74% of these cases (109/147) the cholesterol is less than 200 and in 92% (135/147) the cholesterol is less than 250. In 66% (80/122) of the cases with severe atherosclerosis, the disease developed without evidence of elevated serum cholesterol, diabetes, or hypertension. Blood homocysteine, which has been shown by other studies to be an independent risk factor for atherosclerosis, is recommended for assessing prognosis in these cases.

Carotid artery intimal-medial wall thickening and plasma homocyst(e)ine in asymptomatic adults. The Atherosclerosis Risk in Communities Study.
Malinow MR; Nieto FJ; Szklo M; Chambless LE; Bond G: Circulation (United States), Apr 1993, 87(4) p1107-13

Editor's Comment:
I remember meeting Dr. Malinow in New York City in Dr. Victor Herbert's apartment. This was followed by a great Thai dinner on 59th street. I took the opportunity to discuss Dr. Malinow's homocysteine studies and found him to be a credible, brilliant researcher who was ahead of his time. The following work is a landmark study of great importance.

Dr. Malinow constructed a rather simple, noninvasive study using ultrasound to measure the thickness of the wall of the carotid arteries, the vessels that pipe all of the blood to the head. It is these vessels that when thickened and narrowed lead to strokes and the so-called TIA (transient ischemic attack). The study as presented below shows that if your homocysteine level is high, in the upper twenty percent, the chances for a thickened artery wall are more than three times greater than for someone having a level in the lowest 20%.

Plasma levels of homocysteine are elevated in certain patients with occlusive arterial diseases. We extended these findings to asymptomatic adults. METHODS AND RESULTS. We determined plasma homocysteine levels in 287 pairs of asymptomatic adults. Cases and controls were defined on the basis of intimal-medial thickness of the carotid wall as measured by B-mode ultrasound. Study subjects had no history of atherosclerotic disease and were
selected from a probability sample of 15,800 men and women between 45 and 64 years old. Subjects with thickened intimal-medial carotid walls (cases) had higher plasma homocysteine levels than controls (p 0.001). The odds ratio for having a thickened carotid artery wall was 3.15 (p 0.001) for subjects in the top quintile of plasma homocysteine levels (10.5 mumol/L) compared with those in the bottom quintile (5.88 mumol/L). CONCLUSIONS: The present study as well as observations on the common occurrence of elevated plasma homocysteine levels in patients with occlusive arterial diseases suggest that clinical trials should be conducted to determine whether normalization of hyperhomocysteinemia may prevent progression of atherosclerosis.

**Promotion of vascular smooth muscle cell growth by homocysteine: a link to atherosclerosis.**
Tsai JC; Perrella MA; Yoshizumi M; Hsieh CM; Haber E; Schlegel R; Lee ME: J Proc Natl Acad Sci U S A (United States), Jul 5 1994, 91(14) p6369-73 Cardiovascular Biology Laboratory, Harvard School of Public Health, Boston, MA.

**Editor's Comment:**
This study by Tsai and associates from a prominent laboratory is very provocative. Modern theories of the mechanisms of atherosclerosis propose that an initial toxic event leads to damage of the innermost blood vessel lining cells, the intimal or endothelial cells. After this comes the monocyte scavenger cells and sticky granulocytes (both white blood cells) in an attempt to repair the damage. This is to no avail for the repair mechanism itself becomes a confounding damaging force.

Monocytes change shape as they bore their way into the blood vessel wall and secrete materials that help turn on other cells to grow. They, in concert with other cells, start to imbibe oxidized lipoprotein-cholesterol forming what have been called “foam cells”. The following study points to homocysteine's ability to not only damage endothelial cells (the initial event of atherosclerosis) but also to stimulate the overgrowth of blood vessel smooth muscle cells, a hallmark of atherosclerosis.

Plasma homocysteine levels are elevated in 20-30% of all patients with premature atherosclerosis. Although elevated homocysteine levels have been recognized as an independent risk factor for myocardial infarction and stroke, the mechanism by which these elevated levels cause atherosclerosis is unknown. To understand the role of homocysteine in the pathogenesis of atherosclerosis, we examined the effect of homocysteine on the growth of both vascular smooth muscle cells and endothelial cells at concentrations similar to those observed in clinical studies. As little as 0.1 mM homocysteine caused a 25% increase in DNA synthesis, and homocysteine at 1 mM increased DNA synthesis by 4.5-fold in rat aortic smooth muscle cells (RASMC). In contrast, homocysteine caused a dose-dependent decrease in DNA synthesis in human umbilical vein endothelial cells. Homocysteine increased mRNA levels of cyclin D1 and cyclin A in RASMC by 3- and 15-fold, respectively, indicating that homocysteine induced the mRNA of cyclins important for the reentry of quiescent RASMC into the cell cycle. Furthermore, homocysteine promoted proliferation of quiescent RASMC, an effect markedly amplified by 2% serum. The growth-promoting effect of homocysteine on vascular smooth muscle cells, together with its inhibitory effect on endothelial cell growth, represents an important mechanism to explain homocysteine-induced atherosclerosis.

**Association between plasma homocysteine concentrations and extracranial carotid-artery stenosis.**
Selhub J; Jacques PF; Bostom AG; D’Agostino RB; Wilson PW; Belanger AJ; O’Leary DH; Wolf PA; Schaefer EJ; Rosenberg IH: Comment in: N Engl J Med 1995 Feb 2; 332(5):328-9 Department of Agriculture Human Nutrition Research Center on Aging, Tufts University, Boston, MA 02111.

**Editor's Comment:**
Dr. Selhub has written extensively on homocysteine and atherosclerosis. Here is a study that not only confirms the results of Malinow's work cited above but also correlates high blood levels of homocysteine with decreased ingestion of vitamins B6 and folic acid. The authors were also able to correlate lower serum levels of these vitamins in patients with thickened carotid arteries and high homocysteine.

Later, in another chapter, I will present evidence revealing that it is next to impossible to ingest enough folic acid from a healthy diet to suppress homocysteine to its lowest and safest level. The roles played by vitamin B6 and folic acid are critical and will be explained in detail in the chapter on homocysteine metabolism.

Epidemiological studies have identified hyperhomocysteinemia as a possible risk factor for atherosclerosis. We determined the risk of carotid-artery atherosclerosis in relation to both plasma homocysteine concentrations and nutritional determinants of hyperhomocysteinemia. METHODS. We performed a cross-sectional study of 1041
elderly subjects (418 men and 623 women; age range, 67 to 96 years) from the Framingham Heart Study. We examined the relation between the maximal degree of stenosis of the extracranial carotid arteries (as assessed by ultrasonography) and plasma homocysteine concentrations, as well as plasma concentrations and intakes of vitamins involved in homocysteine metabolism, including folate, vitamin B12, and vitamin B6. The subjects were classified into two categories according to the findings in the more diseased of the two carotid vessels: stenosis of 0 to 24 percent and stenosis of 25 to 100 percent. RESULTS. The prevalence of carotid stenosis of or = 25 percent was 43 percent in the men and 34 percent in the women. The odds ratio for stenosis of or = 25 percent was 2.0 (95 percent confidence interval, 1.4 to 2.9) for subjects with the highest plasma homocysteine concentrations (or = 14.4 mumol per liter) as compared with those with the lowest concentrations (or = 9.1 mumol per liter), after adjustment for sex, age, plasma high-density lipoprotein cholesterol concentration, systolic blood pressure, and smoking status (P 0.001 for trend). Plasma concentrations of folate and pyridoxal-5'-phosphate (the coenzyme form of vitamin B6) and the level of folate intake were inversely associated with carotid-artery stenosis after adjustment for age, sex, and other risk factors. CONCLUSIONS. High plasma homocysteine concentrations and low concentrations of folate and vitamin B6, through their role in homocysteine metabolism, are associated with an increased risk of extracranial carotid-artery stenosis in the elderly.


Editor's Comment:
This study evaluated 21,826 subjects and confirmed the association of high homocysteine levels with heart attack. It also found the absence of a threshold value for homocysteine above which heart attacks occur. This means that those with lower homocysteine levels will have a lower chance of having a heart attack. As we shall discuss in another chapter, therapy with appropriate doses of vitamins will lower serum homocysteine levels by up to 50%.

Several studies have observed high plasma levels of homocysteine among patients with coronary heart disease (CHD). The only prospective study was based on US physicians, and concluded that homocysteine was associated with subsequent myocardial infarction (MI). However, the association was limited to those above a threshold level of homocysteine. METHODS. We conducted a nested case-control study among the 21,826 subjects, aged 12-61 years, who were surveyed in the municipality of Tromso, Norway. Among those free from MI at the screening, 123 later developed CHD. Four controls were selected for each case. RESULTS. Level of homocysteine was higher in cases than in controls (12.7 +/- 4.7 versus 11.3 +/- 3.7 mumol/l (mean +/- SD); P = 0.002). The relative risk for a 4 mumol/l increase in serum homocysteine was 1.41 (95% confidence interval (CI): 1.16-1.71). Adjusting for possible confounders reduced the relative risk to 1.32 (95% CI: 1.05-1.65). There was no threshold level above which serum homocysteine is associated with CHD events. CONCLUSIONS. In the general population serum total homocysteine is an independent risk factor for CHD with no threshold level.