

Letters to the Editor

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Cerebral Atherosclerosis and Coronary Calcification

To the Editor:

We read with great interest the article by Vliegenthart et al¹ concerning an association between stroke and coronary calcification. It is of interest whether coronary calcification is correlated with cerebral atherosclerosis. We also believe that coronary calcification could play a crucial role in the incidence of cerebral infarctions. We would like to show a possible relationship between coronary calcification and cerebral atherosclerosis and then we would like to compare their study and ours.

At the PL Tokyo Health Care Center, 31 646 Japanese subjects (19 901 men and 11 745 women) received physical checkups between April 1, 2001, and March 31, 2002. Among them, brain checkup and helical CT of the chest were done in 1100 subjects (865 men, 245 women). Mean age was 53.8 years (SD, 10.9 years), 54.0 years (SD, 10.8 years) in men and 53.1 years (SD, 11.2 years) in women. Brain MRI and magnetic resonance angiography (MRA) were produced by a 1.5-Tesla superconducting system (Stratis II, Hitachi Medical Co). Axial T1-weighted (repetition time/echo time=400/20 ms) and T2-weighted (repetition time/echo time=4750/120 ms) images were performed on MRI. The slice/gap thickness of the MRI was 6.0/0.5 mm, and the matrix size was 224×256. The number of acquisitions was 2. MRA was applied by 3-dimensional time-of-flight technique. The slice thickness was 0.6 mm, and the matrix size was 140×140. Helical CT was used with CT-W3000 (Hitachi Medical Co). The slice thickness was 5 to 10 mm, and the matrix size was 256×256. Scan areas included the root of aorta through the heart. The total number of slices was 40.

Lacuna was defined as T1-hypointense and T2-hyperintense areas (3 mm< diameter<15 mm). MRA score of atherosclerosis was classified from grade 1 to 4. In grade 1 (normal), blood flow signal intensities were displayed clearly in the A3 segment of the anterior cerebral artery, M3-4 segment of the middle cerebral artery, or P3-4 segment of the posterior cerebral artery. In grade 2 (mild atherosclerosis), blood flow signal intensities were seen equivocally in the distal portion of the A3 segment, M3 segment, or P3 segment. In grade 3 (moderate atherosclerosis), blood flow signal intensities were absent in the distal portion of the A3 segment, distal portion of the M3 segment, or distal portion of P3 segment. Finally, in grade 4 (severe atherosclerosis or obstruction), blood flow signal intensities were absent in the A1 segment of the proximal portion of A3 segment, M1 segment of the proximal portion of the M3 segment, or P1 segment of the proximal portion of P3 segment.

Total atherosclerotic score was calculated as 3 to 12 in the anterior, middle, and posterior cerebral arteries. One experienced neurologist and 2 diagnostic radiologists reviewed brain MRI and MRA. Three experienced diagnostic radiologists reviewed chest CT. Calcium score was determined according to the method of Agatston et al.² Coronary calcification was diagnosed as calcium score >500 and a density of >130 Hounsfield units in the epicardial coronary arteries. One hundred forty-nine subjects (131 men, 18 women) had coronary calcification. The incidence of calcification was 12.5%, 14.2% in men and 6.5% in women. The mean age of the calcification group was 63.9 years (SD, 8.7 years), 63.1 years (SD, 8.6 years) in men and 70.3 years (SD, 6.6 years) in women. That of the noncalcification group was 52.3 years (SD, 10.4 years), 52.4 years (SD, 10.3 years) in men

and 51.9 years (SD, 10.5 years) in women. The frequency of diabetes mellitus (fasting plasma glucose >126 mg/dL or current medication), hypertension (systolic pressure >140 mm Hg, diastolic pressure >90 mm Hg, or current medication), or hypercholesterolemia (total cholesterol >220 mg/dL or current medication) was significantly higher in the coronary calcification group compared with the noncalcification group. Current or history of smoking was also seen more frequently in subjects with coronary calcification. The number of lacunas did not differ between subjects with and without coronary calcification. The MRA score was associated with hypertension and diabetes mellitus. Severity of cerebral atherosclerosis and incidence of coronary calcification were increased by age in our adult subjects. The atherosclerotic score was significantly higher in subjects with coronary calcification than in those without calcification. In logistic regression analysis, there were no statistical associations between coronary calcification and the degree of cerebral atherosclerosis when adjusted for diabetes mellitus, hypertension, hypercholesterolemia, and smoking. An age-adjusted model showed a statistical tendency between coronary calcification and cerebral atherosclerosis in women (odds ratio, 3.7) but not in men (odds ratio, 1.3).

Vliegenthart et al¹ report the association between coronary calcification and stroke history in Rotterdam older inhabitants. We evaluated the degree of cerebral atherosclerosis in individuals with coronary calcification on the basis of the retrospective data of helical CT, brain MRI, and MRA. Our data indicated that subjects with coronary calcification had higher cerebral atherosclerotic score and several cardiovascular risk factors. The calcification subjects were ≈10 years older than the noncalcification subjects. We would like to know the mean age of subjects with calcium scores >500 in the study of Vliegenthart et al. In addition, what is the frequency of hypertension, diabetes mellitus, and hypercholesterolemia and the smoking history in those subjects? Vliegenthart et al suspect the possibility that most of stroke types may consist of lacunar infarctions in their study. Our data of brain checkups show that asymptomatic lacunar infarction occurs in 20% to 30% of healthy senile subjects. We would like to know neuroradiological data if they performed brain CT, MRI, or MRA in subjects with calcium scores >500. The percentage of calcium scores >500 was lower in our study (12.5%) than in the Rotterdam study (27.5%). Vliegenthart et al applied electron-beam CT scan. A severe degree of coronary calcification on conventional helical CT was detected in our subjects. Helical CT is known to be restricted for the quantitative measurement of coronary calcification compared with electron-beam CT.³ The age of subjects also differs between their study and ours. The mean age was 70.8 years (SD, 5.5 years) in the study of Vliegenthart et al¹ and 53.8 years (SD, 10.9 years) in our subjects. A previous report suggests that calcium score increases with age in older adults.⁴

In our study, coronary calcification and cerebral atherosclerosis are associated with age and several cardiovascular risk factors. Age-adjusted logistic analysis discloses a statistical tendency between coronary calcification and cerebral atherosclerosis in our female subjects. Further long-term studies are needed to determine whether coronary calcification is an independent risk factor of stroke in the Japanese population.

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Response

Dr Ikeda reports a positive association between coronary calcification and cerebral atherosclerosis evaluated by MRI and MRA. The severity of cerebral atherosclerosis was related to the presence of a calcium score >500. These findings are in agreement with previously published data from our study¹. We found a graded association between the amount of coronary calcification and the intima-media thickness of the common carotid artery, and an even stronger association with the presence of carotid plaques. The MRI data from Ikeda et al allow assessment of lacunar infarctions, whether symptomatic or asymptomatic. Unfortunately, MRI was not performed in our study, except for those patients who experienced a stroke and subsequently had diagnostic evaluation in hospital.² However, the association between cerebral atherosclerosis and coronary atherosclerosis is interesting and deserves further evaluation.

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Parallel Morning and Evening Surge in Stroke Onset, Blood Pressure, and Physical Activity

To the Editor:

I read with great interest the excellent study by Stergiou et al.¹ It demonstrates what has been predicted by an afternoon increase in blood pressure and heart rate in patients when awakening from a siesta.² Actually, it was shown that the siesta might be a novel risk factor for vascular mortality in elderly subjects.³ Moreover, it was found that this occurs after an afternoon sleep but not when those who practice it just rest in bed without sleeping.⁴

Thus, it corroborates the prediction by Mulcahy et al⁵ that the siesta may in fact be a “snooze induced excitation of triggered sympathetic activity”⁵ rather than a protective activity as usually intuitively perceived.^{6,7}

On the background of the exciting findings of Stergiou et al,¹ it is interesting to speculate that the overlooked and thus as-yet-unidentified habit of the siesta in the agricultural and hot “stroke belt” in the United States may account for the as-yet-unexplained excess of strokes in that area.⁸

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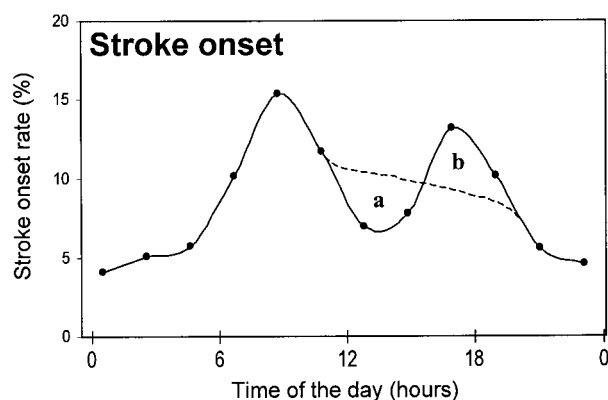
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Response

We thank Dr Bursztyn for his interest in our article. The hypothesis he advances is quite interesting, but the existing evidence is too limited to allow judgment as to whether the siesta habit affects in a beneficial¹ or a detrimental way² the occurrence of cardiovascular events.

It is clear that our study was designed not to address this issue but to investigate the timing of stroke onset within the 24-hour period compared with the diurnal variation in blood pressure and physical activity.³ Nevertheless, one might attempt to compare the afternoon trough with the evening peak in stroke incidence in our study. Thus, a hypothetical diurnal variation curve (the Figure, dotted line) for stroke onset in subjects who do not take a siesta might be added in the figure of the diurnal variation curve observed in our study.³ Comparison of the observed with the hypothetical 24-hour curves suggests that the practice of siesta may be associated with stroke protection (afternoon trough “a,” where the incidence of stroke was reduced during siesta) or with increased risk for stroke (evening peak “b,” where a surge in the incidence of strokes was observed). Whether the excess



Observed diurnal variation curve for stroke onset in all patients in the study (solid line)³ and hypothetical variation curve in those who do not take a siesta (dotted line).

risk for stroke on waking and rising in the evening overcomes the afternoon protection during siesta depends on the curve-fitting parameters.

Perhaps a direct comparison of stroke incidence in patients who regularly take a siesta with those who regularly do not would allow a critical evaluation of the 2 alternative hypotheses.^{1,2} Unfortunately, although the practice of siesta is known to be common in the population included in our study, specific information on this issue for stroke patients was not obtained. Therefore, at present the evidence regarding the effect of siesta on cardiovascular risk is weak.

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Patient Selection for Carotid Angioplasty and Stenting

To the Editor:

We read with interest the article by Dietz et al¹ recently published in *Stroke*. The authors report the clinical results of a consecutive series of 43 patients with symptomatic carotid stenosis treated by angioplasty and stenting. We would like to express concern about the selection of patients for endovascular treatment in this study.

In the title and within the article, the authors emphasize the selection of patients according to the prognostic model of Rothwell et al² for carotid endarterectomy. This model was developed from data gathered in the European Carotid Surgery Trial³ (ECST) to identify the best candidates for surgical treatment as opposed to medical therapy alone. The model included 2 sets of prognostic factors for medical and surgical adverse events and provided a score from 0 to 5 that predicted benefit (≥ 4), no significant benefit (>1 to <4), or potential harm (≤ 1)

of the operation. The conclusion drawn from Rothwell et al was to exclude patients with scores <4 from the operation and to treat them exclusively with prophylactic medical therapy.

The prognostic model was developed from data on patients with 0% to 69% carotid stenosis in the ECST and then tested and validated on 990 ECST patients with 70% to 99% carotid stenosis assigned to surgery (594) or medical therapy alone (396). To the best of our knowledge, the prognostic model has not yet been validated by institutions other than ECST Collaborative Group, especially not by single centers confronted with the selection of individual patients for carotid endarterectomy.

Our concern about the selection procedure chosen by Dietz et al to indicate endovascular treatment of carotid stenosis is that patients with a Rothwell score <4 have been excluded from surgery but not treated medically as would be requested by the model. In fact, based on a prognostic model that has not yet been validated by independent centers, these patients have been denied a therapy, carotid endarterectomy, whose safety is established, only to be exposed to a treatment, angioplasty and stenting, whose safety and efficacy are still in investigation. In our opinion, such a procedure is questionable.

From a scientific point of view, a statistical model developed for a certain treatment cannot automatically be used for another therapy. The 3 surgical risk factors elaborated by Rothwell et al to calculate their prognostic scores were derived from patients treated exclusively with open endarterectomy. They included female sex, peripheral vascular disease, and systolic blood pressure >180 mm Hg. Before the model can be extended to angioplasty and stenting, it has to be proved that these factors keep their prognostic value in patients treated with angioplasty and stenting.

From an ethical point of view, we disapprove of the exclusion of patients from the standard treatment to recruit them for a new therapeutic modality whose safety has not yet been compared with this standard treatment. This is acceptable only in the setting of a prospective randomized trial in which patients are informed about both therapeutic modalities and have the possibility to refuse the proposed new treatment. There are presently several trials running in Europe and the United States that compare both treatments prospectively.⁴ As long as reliable data on the results of these trials are not available, it is recommended to perform primary endovascular therapy of carotid stenosis only in the frame of study protocols. In the study by Dietz et al, patients gave written consent to angioplasty and stenting, but it is not stated whether they were informed about the selection procedure that implied automatic exclusion from the standard treatment, ie, endarterectomy.

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Response

We thank Drs Barth and Bassetti for their critical comments on the article by Dietz et al,¹ but we believe that we can clarify some points they raised. Within the last decade, several investigations provided evidence that factors other than the occurrence of ischemic symptoms and degree of stenosis modulate the risk of stroke and death associated with carotid stenosis. For instance, the occurrence of transient monocular blindness is associated with nearly the half the risk of ipsilateral stroke as that associated with cerebral ischemic symptoms.² Furthermore, the appearance of an irregular/ulcerative plaque surface morphology on angiography was associated with an ≈ 1.8 -fold increased risk of ipsilateral stroke in medically treated patients.^{3,4} After the first ischemic event, the subsequent risk of ipsilateral stroke decreases continuously within the following months.⁵ On the other hand, female sex is associated with a higher risk of perioperative stroke or death in both symptomatic and asymptomatic patients.^{6,7} Thus, confirmed by these independent findings from studies other than the European Carotid Surgery Trial, it seems to be most likely that the risk appraisal function developed by Rothwell and Warlow⁶ is valid and can be used to estimate the individual risk and benefit from endarterectomy in patients with symptomatic high-grade carotid stenosis.

In our study, patient selection was made by an interdisciplinary conference between neurologists, vascular surgeons, and interventional neuroradiologists who selected cases with a suggested increased surgical risk based not only on the above-mentioned Rothwell score but also on a substantial amount of comorbidity or extracranial multivessel disease. All patients were informed about the proven standard therapy of carotid endarterectomy and the new and not finally evaluated character of carotid stenting. The reasons for deviation from standard treatment were explained, as well as the possibility to convert to standard surgical treatment without negative consequences. Perhaps we stressed the role of the Rothwell score too much in the article, but the positive results of our follow-up study with a low periprocedural complication rate and a combined stroke and death rate of 5% within 22 months may support the notion that primary stenting may be a safe and potentially effective treatment option in subgroups of patients with a balanced surgical risk-to-benefit ratio. We still think that it is ethical to investigate and evaluate therapeutic alternatives for this group of patients before we have the final data from a randomized carotid stent trial.

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Nitric Oxide May Contribute to the Long-Term Impairment of Synaptic Transmission After Transient Ischemia

To the Editor:

In their appealing study published in a recent issue of *Stroke*, Bolay et al¹ extend their previous observations² that relatively mild ischemia causes long-term dysfunction of synaptic transmission and now suggest that a likely mechanism for this phenomenon is persistent inhibition of presynaptic signaling and neurotransmitter release. In their convincing experiments, the authors demonstrate that 1-hour middle cerebral artery occlusion causes selective inhibition of synapsin-I phosphorylation in the ischemic penumbra of the rat cortex and propose this as a key reason for the suppression of vesicular neurotransmitter release. Interestingly, the postsynaptic elements of neuronal networks seem to remain intact and continue to respond to exogenously applied glutamate. These findings, together with previous data on ischemia-induced inactivation of Ca^{2+} /calmodulin-dependent protein kinase II³ and protein kinase A,⁴ create a new picture of how relatively mild ischemia or transient ischemic attack may cause long-term impairment of brain function despite survival of neuronal cells.

I agree with the authors that phosphorylation defects likely contribute to the long-lasting suppression of synaptic transmission. However, I would like to attract the attention of the authors and readers to an additional possible mechanism of synaptic transmission impairment. It is a well-known fact that ischemia upregulates production of the free radical nitric oxide (NO).⁵ In the healthy brain, NO serves as a vasorelaxant and neuromodulator. In contrast, under pathological conditions, neuronally derived NO and several related nitrogen reactive species play major roles in mediating brain damage.^{5,6} One such pathological NO product, peroxynitrite (ONOO^-), is formed in the reaction of NO with the superoxide radical.⁷ ONOO^- oxidizes or modifies many cellular components, including proteins and DNA,⁷ and has been quantitatively linked to the degree of ischemic brain damage.^{8,9} In proteins, ONOO^- nitrates tyrosine residues and nitrosates cysteine amino acid moieties, potentially perturbing enzymatic functions.⁷ Synaptic vesicle membrane fusion and exocytotic neurotransmitter release involve an *N*-ethylmaleimide-sensitive ATPase that is especially prone to SH-group oxidation.^{10,11} Recently, my colleagues and I have found that nitric oxide donors potently and selectively suppress vesicular GABA release in brain synaptosomes, while marginally affecting other types of GABA transport.¹² The effects of the NO donors coincided with the oxidation of intrasynaptosomal SH groups and were mimicked by *N*-ethylmaleimide. Therefore, under pathological conditions, NO-dependent disruption of synaptic fusion may work in parallel or in sequence with defects in synapsin-I phosphorylation, as described by Bolay et al. This alternative mechanism deserves further experimental exploration.

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Response

We thank Dr Mongin for his interest in our study and his comments. Dr Mongin proposes an intriguing mechanism that may contribute to persistent synaptic transmission defect seen after transient cerebral ischemia.¹ He and his colleagues demonstrated that NO donors selectively suppressed GABA release in synaptosomes and that this effect was correlated with their ability to oxidize intrasynaptosomal SH groups.² Based on these in vitro data, the author suggests that N-ethylmaleimide-sensitive ATPase, a key enzyme for the fusion step of vesicular exocytosis, might be involved in ischemia-induced impairment of neurotransmitter release because this enzyme is particularly prone to SH-group oxidation by peroxynitrite. The idea is appealing and may provide an additional mechanism that may disrupt neurotransmitter release after an ischemic insult. During ischemia/reperfusion, a considerable amount of peroxynitrite is generated in the penumbral cortex,³ where we detected the synaptic transmission defect.^{1,4} However, it should be noted that neurons in the peri-infarct area are reversibly injured and peroxynitrite toxicity may be reversed by protein denitrosylation after reperfusion. Therefore, peroxynitrite-induced protein modifications may not be sustained. This interesting point may be clarified in future experiments.

Supporting Dr Mongin's view, several studies suggest that peroxynitrite may biochemically modify presynaptic proteins involved in transmitter release including SNARE proteins.^{2,5–7} Even small changes in the formation of SNARE complex and other vesicular proteins could have significant consequences for

vesicle docking and fusion.⁸ Di Stasi and colleagues⁷ demonstrated that peroxynitrite stimulated vesicle exocytosis and induced glutamate release from synaptosomes through nitration of SNAP25 and Munc-18. Peroxynitrite seems to exert opposing effects on release of various neurotransmitters in that it inhibits ACh synthesis⁶ and GABA release,² whereas it stimulates excitatory amino acid release.^{5,7} It would be interesting to know whether NO/peroxynitrite impairs N-ethylmaleimide-sensitive ATPase differentially between inhibitory and excitatory synapses, because selective suppression of GABA release may account for the ischemia-induced cortical hyperexcitability.^{9–12}

As we emphasized before,¹ mechanisms of failure of transmitter release are likely to be complex after an ischemic insult; swelling and depolarization of the presynaptic membrane may adversely affect evoked transmitter release. Synaptic terminals may suffer from significant free radical damage due to a high rate of oxidative phosphorylation and Ca²⁺ shuttling across the mitochondrial membrane during ischemia.¹³ Oxidative and nitrosative stress may damage several macromolecules in synaptic boutons. Mitochondria of synaptic origin have been reported to be much more sensitive to inhibition of complex I and to depletion of glutathione than are nonsynaptosomal mitochondria.¹⁴ Abrupt decrease in ATP synthesis and respiration rate occurred when complex I activity was inhibited by 25% in synaptic mitochondria, whereas this threshold was found to be 72% in nonsynaptic mitochondria.¹⁴ Therefore, mitochondria in presynaptic terminals may be selectively damaged despite recovery of oxidative phosphorylation in other cellular compartments after recirculation. In that case, a generalized deficiency in phosphorylation of presynaptic proteins is expected in addition to phosphorylation defects induced by selective inactivation of some kinases. However, after a mild injury, most of these perturbations may quickly recover, whereas some of them remain dysfunctional and cause long-lasting disturbances in transmitter release. In our study, we detected the deficiency in synapsin-I phosphorylation as one of the possible mechanisms; certainly, several other mechanisms may be identified in future studies, as suggested by Dr Mongin.

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