Correspondence

Plasma homocysteine and MTHFR C677T genotype in levodopa-treated patients with PD

To the Editor: The paper by Yasui et al.[1] showing elevated plasma homocysteine levels in levodopa-treated patients with PD is interesting and confirms other reports[2] and our unpublished findings (table). It also shows that patients with the TT-genotype of the C677T polymorphism of methylenetetrahydrofolate reductase (MTHFR) have especially high homocysteine levels. These findings have to be related to the metabolism of levodopa and methyl groups and may have importance in the future treatment of PD with levodopa.

Homocysteine is the demethylated derivative of methionine and is only formed in numerous transmethylation reactions (e.g., catecholamine *O*-methyltransferase [COMT]) that use S-adenosylmethionine (SAM) as the methyl donor.[3] Most administered levodopa is COMT-methylated to 3*O*-methyldopa and the methylation of 1000 mg of levodopa consumes 5 mMol of SAM methyls and produces 5 mMol of S-adenosylhomocysteine (SAH), which then is deadenosylated to homocysteine. In the study of Yasui et al., the average levodopa dose was 1500 mg. Therefore, it is most likely that levodopa treatment contributed to hyperhomocysteinemia in their patients with PD.

In this context it is important to note that humans normally consume about 15 mMol/day of SAM methyls for methylation of endogenous compounds such as phospholipids, DNA, myeline, and proteins. About 10 mMol of these methyls are supplied from food, mainly as methionine and choline, and about 5 mMol are de novo synthesized in one-carbon (folate) metabolism and are transferred by a vitamin B12-dependent reaction to homocysteine, again forming methionine and SAM.[4]

As we can assume that levodopa-treated PD patients do not increase their intake of labile methyls, they must de novo synthesize all their extra methyls consumed for levodopa methylation. In the above 1000 mg example, the patient must increase his or her de novo synthesis of methyls by about 100%. If methyl neogenesis is hampered by poor folate intake, especially if the patient is a TT-homozygote for C677T/MTHFR polymorphism (about 12% of the Caucasian population), or has a low vitamin B12 level, production of SAM methyls may not be sufficient for both methylation of endogenous compounds and levodopa. This may lead to hypomethylation of essential neural components, and might explain the neuropsychiatric symptoms in folate and vitamin B12 deficiency.[5] This mechanism may contribute to side effects of levodopa. If this is the case, early combination of levodopa with COMT inhibitors (tolcapone, entacapone) would prove to be beneficial. Moreover, studies on the relationship between complications to levodopa therapy in PD and homocysteine, folate, vitamin B12, and C677T/MTHFR genotypes are warranted.

Lars Brattström, MD, PhD, *Kalmar, Sweden*

To the Editor: Yasui et al. found significantly elevated plasma homocysteine levels in 90 levodopa-treated parkinsonian patients with a clear but not significant increase of homocysteine occurring in the homozygous MTHFR C677T genotype.[1] We have performed a similar study, but we found more distinct results. MTHFR C677T genotypes (TT = homozygous; CT = heterozygous; CC = wild-type) were determined by PCR in 82 long-term levodopa-treated patients (age 63.1 ± 10.7 years [mean ± SD]; women/men = 25/57; Hoehn & Yahr scale 2.8 ± 0.9; duration of disease 6.3 ± 5.7 years; daily levodopa dosage 412.6 ± 253.6 mg with no significant difference between the TT, CT, and CC groups) and 39 controls (age 61.5 ± 10.8 years; women/men = 14/25). Standardized measurement of total plasma levels with high performance liquid chromatography was only performed in subjects with no metabolic disturbances such as diabetes mellitus, hypertension, reduced levels of vitamine B6, cobalamine, or folic acid or neurologic diseases other than PD.[6] Each participant fasted and was off any medication for at least 12 hours before blood samples were taken in the morning. Thus we avoided impact of acute levodopa/DDI intake. Patients gave informed consent and the local ethical committee approved this study. We confirm previous results with significantly increased homocysteine levels in parkinsonian patients compared with controls (17.3 ± 9.1 versus 13.0 ± 6.2 μmol/L [mean ± SD]; $p < 0.001$, Mann-Whitney *U*-test).[1] But we found augmented homocysteine levels in TT (n = 12; 22.0 ± 8.5 μmol/L; $p < 0.05$) and CT (n = 36; 18.9 ± 10.9 μmol/L; $p < 0.01$) allele wearing parkinsonian patients compared with the ones with the CC allele (n = 34; 13.9 ± 5.6 μmol/L). We suggest MTHFR genotyping before initiation of levodopa treatment and homocysteine monitoring during levodopa long-term treatment in patients with the TT and CT allele. This may reduce occurrence of increased mortality from cardiovascular disease and stroke, both of which are increased in treated parkinsonian patients.[7,8]

Table *Plasma homocysteine concentration (mean ± SD) in levodopa-treated patients with PD*

Characteristics	Patients	Control subjects
No.	54	132
Men/women	34/20	73/59
Age, y	68 ± 9.1	71.8 ± 4.9
Homocysteine, μmol/L	17.3 ± 5.2	13.1 ± 3.6*

* $p < 0.001$.

Wilfried Kuhn, MD, Thomas Hummel, MD, Dirk Woitalla, MD, Thomas Müller, MD, *Bochum, Germany*

Reply from the Authors: We thank Dr. Brattström for his comments on our paper. We agree that elevated plasma homocysteine levels in patients with PD may be related to the metabolism of levodopa and methyl groups such as SAM and SAH. However, in our paper, 1500 mg of levodopa was equivalent to 300 mg levodopa/carbidopa,[9] as we adjusted the daily levodopa dose in levodopa-treated patients. We believe that the dose of levodopa in our patients is not as likely to induce hyperhomocysteinemia as Dr. Brattström suggests. The main point of our paper was that MTHFR genotype and folate levels also influence homocysteine levels in patients with PD. Although there is no doubt that the combination of levodopa with COMT inhibitor is beneficial, COMT inhibitor has not been available for treatment of patients with PD in Japan. It may be interesting to learn whether SAM levels are correlated to neuropsychiatric symptoms and fluctuation of symptoms because hyperactivity of COMT induces low SAM levels and low levodopa levels. We are studying whether SAM and SAH levels are correlated to such symptoms in patients with PD.

We also appreciate the comments of Kuhn et al. We believe it is reasonable that plasma homocysteine level is elevated in PD not only with TT genotype but also with CT genotype of MTHFR. Our further study for leukocyte MTHFR enzyme activity by the method of Kang[10] shows, in fact, that MTHFR activity in CT genotype patients (n = 30, 8.6 ± 2.8 nmol HCHO/mg protein/h [mean ± SD]) was lower than that of CC genotype (n = 24, 13.1 ± 4.1 nmol HCHO/mg protein/h; $p < 0.0001$), and that of TT genotype (n = 15, 3.9 ± 1.4 nmol HCHO/mg protein/h; $p < 0.0001$, one-way ANOVA followed by Fisher's PLSD) was the lowest. Our additional data coincide with their results. Therefore, their data and our new data firmly support the conclusion that MTHFR genotype is a significant factor for hyperhomocysteinemia in levodopa-treated patients with PD.

K. Yasui, MD, H. Kowa, MD, PhD, K. Nakaso, MD, PhD, T. Takeshima, MD, PhD, K. Nakashima, MD, PhD, *Yonago, Japan*

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FLAIR MRI in sporadic Creutzfeldt–Jakob disease

To the Editor: Vrancken et al. report in their discussion of MRI in Creutzfeldt–Jakob disease (CJD) that variant CJD cannot be differentiated from sporadic CJD by means of MRI.[1] However, some early cases of variant CJD were reported to have bilateral posterior thalamic (pulvinar) high signal on MRI and we suggested that this might be a specific finding.[2] Our subsequent study showed that 28 of 36 (78%) pathologically confirmed cases of variant CJD demonstrated high signal predominantly in the thalamus (in particular the pulvinar) on T2-weighted or proton density–weighted MRI or both, an abnormality we have called the pulvinar sign (see figure, A).[3] Our control group consisted of 57 patients, 32 of whom had a diagnosis of definite or probable sporadic CJD. None of the controls demonstrated the pulvinar sign. Quantitative analysis of MRI signal intensity in variant CJD has supported these findings.[4] Diagnostic criteria for a clinically probable case of variant CJD have now been published, and these incorporate the pulvinar sign.[5]

We have now reviewed MRI scans from a total of 52 cases of pathologically confirmed variant CJD, and identified the pulvinar sign in 41 (79%). We have not found any false positive cases among the patients referred to us.

Vrancken et al. suggests that fluid-attenuated inversion recovery (FLAIR) MRI may have particular diagnostic value in patients with sporadic CJD. Our findings argue that this may also be true for variant CJD. FLAIR images were performed in nine variant CJD cases with the pulvinar sign and in all of these the high signal abnormality was most marked using this sequence (see figure, B).

M. Zeidler, MRCP, D.A. Collie, FRCR, M.A. Macleod, MRCP, R.J. Sellar, FRCR, R. Knight, FRCPE, *Edinburgh, UK*

Reply from the Authors: We are grateful for the valuable addition to our paper by Zeidler et al. Their description of the pulvinar sign on MRI in variant CJD[2-4] does indeed appear to be a promising diagnostic finding. We agree that variant CJD may be differentiated from sporadic CJD when a pulvinar sign is found on MRI. The fact that this abnormality appears to be best visualized on FLAIR MRI supports our argument in favor of this imaging tool in any patient suspected of having CJD.

A.F.J.E. Vrancken, MD, C.J.M. Frijns, MD, L.M.P. Ramos, MD, *Utrecht, the Netherlands*

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Multiple sclerosis and antecedent infections: A case-control study

To the Editor: In a recent paper, Marrie et al.[1] used a case-control methodology to investigate a possible relationship between MS and infectious mononucleosis (IM). The study involved 225 subjects with MS and 900 controls matched for age, sex, and physician practice, with all subjects being from the United Kingdom. Marrie et al., on the basis of a significantly higher rate of IM in cases (5 of 225) than in controls (6 of 900), interpreted that contracting IM is a risk factor for MS.

I believe that their data are not sufficient to draw such a conclusion. Infection with Epstein-Barr virus (EBV), regardless of timing, is a putative risk factor for MS because almost all persons with MS have been previously infected with EBV.[2,3] Experimental work has revealed that EBV could theoretically participate in MS

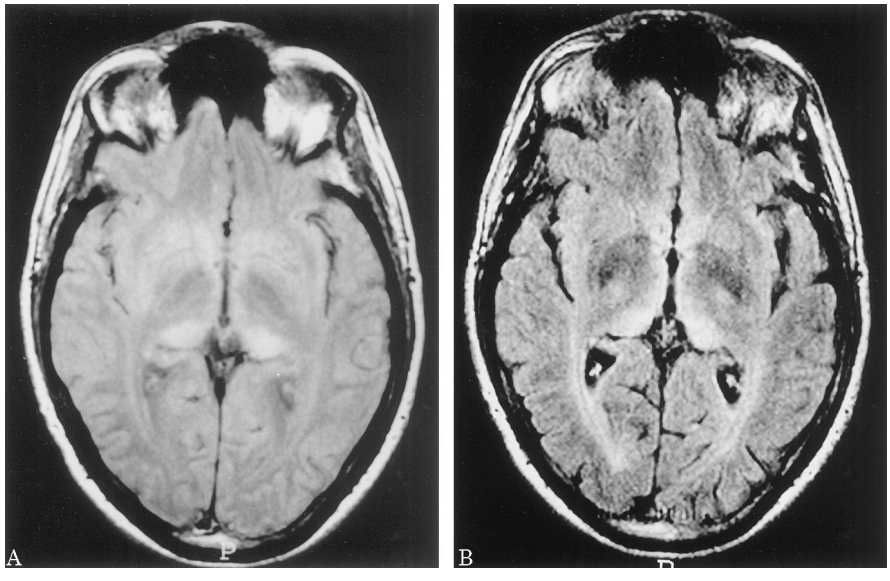

Figure. Axial MRI from a patient with pathologically confirmed variant Creutzfeldt–Jakob disease shows high signal from the pulvinar of the thalamus (arrowheads). (A) proton density–weighted image; (B) fluid-attenuated inversion recovery image.

etiology by providing one or more molecular mimics of myelin antigens.[4,5] Thus, a previous infection with EBV, regardless of an early or late IM timing of the infection, is enough to explain the occurrence of MS in the cases. A standard case-control study cannot provide further insight into specifics of an EBV infection for affecting MS risk.

To investigate whether timing is a significant part of the EBV risk factor, it would be necessary to have controls and cases who are all genetically susceptible to MS and had also all been previously infected with EBV. Then, and only then, could a worthwhile conclusion be drawn regarding the potential for a late infection IM to be a risk factor for MS. Given that it is most likely that the vast majority of the controls (estimated 99+%) in the Marrie et al. study are not genetically susceptible to MS, a comparison of rates of IM between the cases and controls adds nothing to the problem of whether a late infection with EBV is an additional risk factor. Finally, even if a properly controlled future study reveals that IM is indeed a risk factor for MS, the fact that Marrie et al. found that only 2% of the cases in their study had IM before MS onset suggests that IM would be an exceedingly minor factor at best.

Ashton F. Embry, PhD, *Calgary, Alberta, Canada*

Reply from the Authors: Embry has argued that case control methodology is inappropriate for investigating the role of timing of EBV infection as a putative risk factor for the onset of MS, partly because most studies indicate that the majority of the population has been exposed to EBV infection by adulthood. We disagree with this criticism of case control methodology. By examining exposure to a putative risk factor during different time periods before onset of disease, it is certainly possible to obtain useful information even if all cases and controls had been exposed. Indeed, the consideration of a history of exposure during a time period that is not etiologically relevant can lead to the attenuation of the effect estimate.[6] Thus careful attention to the timing of the exposure relative to onset is critical.

In our study we demonstrated an association between IM and subsequent risk of MS (OR = 5.5 [95% CI 1.5–19.7]), with an increased risk associated with IM occurring after age 17 years (OR = 6.0 [95% CI 1.4–25.4]).[1] This suggests that late infection, not simply EBV infection, is a risk factor for MS. This is not a definitive study, as we stated in our article, but it adds to the current body of evidence available.

Embry also suggests that establishing EBV as a risk factor for MS is of minimal importance, as it would explain only a small proportion of MS cases. It is generally accepted that MS is a disease with a multifactorial etiology; thus, we agree with Embry in terms of the ability of EBV exposure to account for a large proportion of MS cases. However, by adding to the body of evidence about the role of EBV in the etiology of MS, we set the stage for larger, more focused epidemiologic research that will include rigorous study of only those factors for which there is strong basic science and epidemiologic evidence.[7]

R.A. Marrie, MD, C. Wolfson, PhD, M.C.J.M. Sturkenboom, PhD, O. Gout, MD, O. Heinzlef, MD, E. Roullet, MD, L. Abenhaim, MD, MSc, ScD, *Montreal, Quebec, Canada*

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Corrections

Past adult lead exposure is associated with longitudinal decline in cognitive function
In the article "Past adult lead exposure is associated with longitudinal decline in cognitive function" by Schwartz et al. (*Neurology* 2000;55:1144–1150), the word "causal" was mistyped as "casual." The sentence that appears on the top of page 1149 should read: "The strength and consistency of this evidence supports a causal relation between past occupational lead exposure and prospective decline in cognitive function." The author apologizes for this error.

Ataxic Guillain–Barré syndrome with anti-GQIb antibody: Relation to Miller Fisher syndrome
In the correspondence "Ataxic Guillain–Barré syndrome with anti-GQIb antibody: Relation to Miller Fisher syndrome" by Derakhshan (*Neurology* 2000;55:1419–1420), the word "myelopathy" was mistyped as "myelinopathy" in the last sentence of the Letter to the Editor. The author apologizes for this error.