Letters to the editor

Factor V Leiden mutation and Japanese NIDDM

Dear Sir,

Activated protein C resistance, associated with a point mutation in the coagulation factor V gene (Arg506Gln), is the most common defect that is found in venous thromboembolic disorders [1]. This mutation, also known as the Leiden mutation, changes Arg506 to Gln, disrupting the activated protein C cleavage site. Previous reports indicate that this mutation is important for an inherited risk for venous thrombosis [2]. However, its roles in the pathogenesis of non-vascular disorders remain unclear. A recent report [3] showed an association of the Arg506Gln mutation with non-insulin-dependent diabetes mellitus (NIDDM), which suggests this mutation is important not only in the case of rare deep vein thrombosis but also in the more prevalent and important disease of NIDDM. Diabetes has been reported to be associated with coagulation and thromboembolic disorders [4, 5] and with various vascular deteriorations [6, 7]. Another recent report [8] suggests that activated protein C sensitivity is decreased in patients with IDDM compared with non-diabetic control subjects. If the factor V Leiden mutation is associated with NIDDM, many of the vascular complications of NIDDM may be partly due to this mutation. The previous report [3], however, was based on an observation of Italian patients with NIDDM; whether this mutation is associated with NIDDM from other ethnic groups remains unclear. To confirm a role for factor V mutation in NIDDM, we investigated whether this mutation is associated with Japanese NIDDM patients. An association of the Leiden mutation with NIDDM in the Japanese has not been investigated previously to our knowledge.

In total, 314 randomly selected unrelated Japanese patients with NIDDM (184 males/130 females, 57.7 ± 11.4 (mean $\pm$ SD) years, range: 31–87 years) and 139 non-diabetic control subjects (78 males/61 females, 49.5 ± 14.0 years, 40–82 years) were recruited from outpatient clinics of the University of Tsukuba Hospital and its affiliated hospitals with informed

consent. The diagnosis of NIDDM was based on WHO criteria [9]. The presence of the mutation was determined by amplifying the fragments encompassing the factor V Leiden mutation by polymerase chain reaction (PCR), being digested with restriction endonuclease Mln I and separated on 7% polyacrylamide gels. For amplifications we used: sense primer, 5'-TGCCCAGTGCTTAACAAGACCA-3', anti-sense primer, 5'-CTTGAAGGAAATGCCCCATTA-3'. The PCR conditions were: 35 cycles of denaturation at 91°C for 40 s, annealing at 55°C for 40 s, and extension at 71°C for 2 min. None of the NIDDM patients or non-diabetic control subjects analysed carried this mutation (Table 1), suggesting an absence of an association of this mutation with NIDDM in Japanese. This mutation seems to be rarely found in the Japanese population. Marked ethnic differences in genetic predisposition to NIDDM have been reported [10, 11]. Unlike its association with Italian patients with NIDDM, the factor V mutation does not seem to be associated with NIDDM in the Japanese population. A study on the distribution of the mutation indicates that this mutation is more common in populations in Europe or in Asia Minor [12]. Investigations of more patients from many populations including those from Europe and Asia Minor seem to be necessary to confirm whether this mutation is really associated with NIDDM. This seems to be important because the mutation could help to explain the increased cardiovascular risk associated with NIDDM in some ethnic groups.

Yours sincerely,
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Table 1. The prevalence of the Factor V Leiden mutation in Japanese diabetic and non-diabetic subjects

	<i>n</i>	Males/Females	Age (years)	Age range (years)	Leiden mutation
NIDDM patients	314	184/130	57.7 ± 11.4	31–87	0
Non-diabetic subjects	139	78/61	49.5 ± 14.0	40–82	0

Data are mean ± SD

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Anti-ganglioside GM1 antibody and distal symmetric “diabetic polyneuropathy” with dominant motor features

Dear Sir,

The prevailing hypothesis of the pathogenesis of chronic diabetic complications is that hyperglycaemia initiates complex metabolic perturbations, which result in pathologic changes in various organs, including the peripheral nervous system. This was amply supported by the Diabetes Control and Complications Trial (DCCT) wherein near normoglycaemia reduced the risk for development of complications [1]. However, once neuropathy is established it proceeds inexorably and cannot be reversed by near-normal glycaemia, for example that achieved by pancreatic transplantation [2, 3]. This could be explained by the fact that neuropathy is a heterogeneous disorder with a multiplicity of aetiologies acting through vascular, immunologic, and metabolic factors [4, 5], as well as deficient neurotrophic support that may also be involved in the progression of neurologic deficit in diabetic patients [4–6].

Activation of immune effectors may be another mechanism capable of causing neuropathy or perpetuating it, once established, by a variety of mechanisms. Immunopathologic studies have revealed the presence of antibodies (Ab) and complement in the peripheral nerves of patients with distal symmetric polyneuropathy (DSPN), as well as increased numbers of cytotoxic T lymphocytes [7, 8]. These findings support the concept of immune system involvement in the pathogenesis of diabetic neuropathy, in addition to the metabolic and microvascular abnormalities.

To further investigate immune phenomena in diabetic neuropathy, we screened a group of 67 consecutive patients with both diabetes and neuropathy for antibodies

against various neuronal antigens, including sulfoglucuronyl paragloboside, sulfatide, neuronal nuclear antigen, myelin associated glycoprotein and the gangliosides: GM1; asialo

Table 1. Demographic characteristics, neuropathy scores (NSS, sensory NDS and motor NDS or MRCS) and quantitative sensory testing in 14 GM1 Ab positive and 53 Ab negative diabetic patients with DSPN

	DSPN	DSPN with anti-GM1 antibodies
<i>n</i>	53	14
Age (years)	54.6 ± 2.1	53 ± 4.3
male/female	32 : 21	6 : 8
IDDM : NIDDM	16 : 37	3 : 11
Duration of diabetes (years)	13.0 ± 1.3	15.2 ± 2.6
Duration of neuropathy (months)	40.2 ± 7.7	46.0 ± 13.1
HbA _{1c} (%)	8.1 ± 0.5	7.9 ± 1.0
Height (cm)	171 ± 1.5	167 ± 3.3
Weight (kg)	86.8 ± 3.0	75 ± 4.9
Body mass index (kg/m ²)	29.4 ± 1.0	25.8 ± 1.8
NSS	3.2 ± 0.3	4.2 ± 0.6
Sensory NDS	23.4 ± 2.3	31.3 ± 4.0
MRCS	2.0 ± 0.8	6.5 ± 1.5 ^a
Vibration threshold (VU)	11.5 ± 1.7	11.9 ± 2.9
Cold detection threshold (°C)	6.1 ± 2.1	9.0 ± 3.3
Warm detection threshold (°C)	8.7 ± 1.8	10.9 ± 1.5
Cold pain threshold (°C)	26.0 ± 4.7	28.0 ± 1.9
Heat pain threshold (°C)	13.5 ± 1.8	14.4 ± 3.4

Quantitative sensory testing included vibration threshold, cold and warm detection thresholds, and thermal pain thresholds
Data are mean ± SEM

VU, Vibration units; NSS, neuropathy symptom score; NDS, neuropathy disability score; MRCS, Medical Research Council Scale or motor neuropathy disability score

^a *p* < 0.05