Mutation of SBDS and SH2D1A is not associated with aplastic anemia in Japanese children

During the last decade, the genetic basis of inherited bone marrow failure syndromes has been identified. Recently, genetic factors predisposing to aplastic anemia (AA) have been found in a few patients with apparently acquired AA. Here we present the genetic analysis of SBDS and SH2D1A in Japanese children with AA.

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The genetic basis of inherited bone marrow failure syndromes (IBMFS) has been clarified in recent years. A study also suggests that cryptic IBFFS may be hidden behind what is apparently acquired aplastic anemia (AA).[1] One characteristic of white blood cells in AA is a short telomere, found in one third of patients with AA.[2] Telomere is known to be involved in aging, oncogenesis and apoptosis and it is maintained by telomerase complex. Germ line mutations in genes encoding the telomerase complex, such as *TERC* and *TERT*, have been identified in less than 10% of patients with AA.[3-4] We also found mutations in *TERT* in 2 out of 96 Japanese children with AA, neither of whom had the clinical characteristics of dyskeratosis congenita.[5] Because telomere shorting in white blood cells has also been identified in patients with Shwachman-Diamond syndrome (SDS),[6] *SBDS* is one of the candidates for association with AA. A recent study showed a heterozygous mutation of *SBDS*, 258+2T>C, in 4 out of 91 patients with apparently acquired AA.[7] In addition, a mutation of *SH2D1A* causes X-linked lymphoproliferative syndrome which is associated with fatal Epstein-Barr virus (EBV) infection and AA.[8] Given that a small population of patients with primary EBV infection develops AA, SH2D1A is also a candidate for association with AA.

We analyzed *SBDS* and *SH2D1A* in 96 Japanese children with AA. The diagnosis of SDS was clinically excluded because findings such as exocrine pancreatic insufficiency and metaphyseal chondrodysplasia were not presented. EBV infection was also excluded because features of the infectious mononucleosis, such as lymphadenopathy and the appearance of atypical lymphocyte in the peripheral blood, were absent. Documented informed consent was obtained from the parents of each patient according to a protocol approved by the Institutional Review Board at Nagoya University, Graduate School of Medicine. We collected blood samples from 96 unrelated children with AA (54 boys and 42 girls) for analysis of *SBDS* and *SH2D1A*. The median age at onset of disease was 7 years (0-16 years). Family history was negative for all patients. Twenty-two patients had very severe AA, 39 had severe AA and 35 had moderate AA. The etiologies of AA were as follows: hepatitis in 7 patients, viral infection in 1 patient and idiopathic in 88 patients. Genomic DNA was isolated from the peripheral blood mononuclear cells and then amplified by PCR using primers, as previously described.[9,10] The coding regions of these genes were fully sequenced, including exon-intron boundaries. Reference genomic sequences obtained from the NCBI Genome Database (*ENTREZ SNP*). No mutations in *SBDS* were detected, while two silent nucleotide changes in exon 2 of SBDS, 141 C>T and 201 A>G, were found in 2 and 1 patient respectively. Neither mutation nor polymorphism of *SH2D1A* was identified. Nakashima *et al*. identified 141 C>T in 1 out of 6 families with SDS and in 5 out of 70 Japanese controls.[10] 201 A>G was found in 2 out of 6 families with SDS, but not in 70 healthy controls. The single nucleotide polymorphism (SNP) found in patients with AA and normal controls is not caused by the gene conversion of *SBDS* with *SBDSP*. We, therefore, compared the incidence of SNPs in patients with AA with that in normal controls. As a result, the frequency of 141 C>T in our study (2/192) was lower than that observed by Nakashima *et al*. (5/140). The frequency of 201 A>G in our study (1/192) was low, as observed by Nakashima *et al*. (0/140). Calado *et al*. reported similar results.[7] Taken together, SNP neither influences the likelihood of AA. In addition, the incidence of 201 A>G is much lower in the Japanese population compared with that in western countries.[7] This result shows the genetic heterogeneity among different ethnic groups. These findings suggest that the gene conversion or SNPs of *SDBS* and *SH2D1A* mutation in the pathogenesis of acquired AA might be limited to specific ethnic groups. Taken together our previous finding,[5] Altogether, given our previous findings,[5] we conclude that aberrations in IBMFS-associated genes rarely contribute to the pathogenesis of AA in Japanese children.

All authors contributed to the design and development of the study as well as the interpretation of data. All approved the final version of the manuscript. The authors declare that they have no potential conflicts of interest nor any financial support.

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