The S65C mutation in Spain. Implications for iron overload screening

Hereditary hemochromatosis is related to mutations of the HFE gene. The role of the S65C mutation of this gene was evaluated in a Spanish population, consisting of 100 controls and 41 patients who had resulted positive to screening for iron overload. Only one patient was heterozygous for the S65C mutation, so the S65C mutation is infrequent in our area. Nevertheless, it is advisable to search for this mutation in cases with iron overload and heterozygosity for the C282Y or H63D mutations of the HFE gene.

Sir, Hemochromatosis is a hereditary disorder that is common in people of European descent. Most cases are attributable to homozygosity for the mutation C282Y in the HFE gene, or compound heterozygosity for the C282Y and the H63D mutations. However, a few individuals with clinical hemochromatosis lack these phenotypes. Other mutations of the HFE gene could account for some of these cases. In this regard, a third mutation in the HFE coding region (exon 2, nt 193A → T, S65C) has been described in hemochromatosis patients (compound heterozygotes) and controls in Brittany and the USA. The proximity of the S65C and H63D substitutions suggests that the former plays a role in hemochromatosis. The S65C mutation was evaluated in 141 individuals in whom C282Y and H63D homozygosities and compound heterozygosity had been ruled out. The S65C mutation was studied with a HinfI digest using the conditions for the H63D analysis. These subjects included 100 control subjects (selected from a large group so that all HFE mutations would be represented) and 41 patients who were positive to screening tests for iron overload (transferrin saturation exceeding 50% and serum ferritin exceeding 450 µg/L) (Table 1). In the latter group, 2 patients had iron overload (measured in a liver biopsy) and 33 patients had hepatitis C virus infection: their characteristics have been reported elsewhere. None of the cases in the control group had the S65C mutation and only one patient out of 41 who screened positive for iron overload was heterozygous for the S65C mutation (genotype CC/HH/SC). When both groups were combined, the estimated allelic frequency was 0.36±0.03%. The frequency of the S65C substitution was lower in our population than in others from different areas (5.5% in Utah, 1.95% in Brittany and 0.6% in Alabama), in contrast to the frequency of the H63D mutation (15-20%), which was higher. It should be pointed out that the frequency observed in our study is probably an overestimation given that we included a number of patients with positive iron overload screening tests. The role of this mutation in iron abnormalities of hepatitis C virus infected patients is negligible. However, a number of arguments justify the evaluation of this substitution in patients in whom the C282Y and H63D mutations do not account for the iron overload. First, the laboratory analysis of this mutation is fairly simple. Second, some compound heterozygotes for S65C have iron overload. Finally, using the crystal structure of the HFE molecule, the position of the S65C mutation within the α1 domain suggests that it has a role in the function of the HFE protein.

In conclusion, the S65C mutation of the HFE gene is very rare in our area of Spain (Northeast). Nevertheless, its evaluation is important when iron overload is demonstrated and heterozygosity for the other mutations (C282Y and H63D) is present.

Angel F. Remacha,* M. Jesús Barceló,* M. Pilar Sardà,* Irene Blesa,* Albert Altés,* Montserrat Baiget*
* Hematology, Genetics and Gastroenterology Departments, Hospital de Sant Pau, Barcelona, Spain

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Correspondence
Angel F. Remacha M.D., Hematology Department, Hospital de Sant Pau, Avda Padre Claret 167. Barcelona 08025 Spain. Phone: international +34.93.2919290 – Fax: international +34.93.2919192 – E-mail: 2107@hsp.santpau.es

References
Differences in phenotype, growth factor requirements, pattern of expression of adhesion molecules and rate of apoptosis displayed by three new myeloid sister leukemic cell lines

We established three new human myeloid cell lines from one patient, in the presence of granulocyte-macrophage colony-stimulating factor (UPM1-GM), interleukin-3 (UPM1-IL-3) or without exogenous growth factors (UPM1). The 3 lines were characterized by phenotypic, genotypic and functional studies. These cell lines may provide useful tools to study different aspects of leukemia cell biology.

Three sister cell lines were established from a bone marrow sample (80% myeloblasts), collected in a resistant phase after 2 courses of induction chemotherapy (mitoxantrone/cytarabine) of an acute myeloid leukemia, FAB type M4Eo. The cells were initially cultured in RPMI 1640 with 20% fetal bovine serum. Three different cell lines were established from the sample, depending on the exogenous stimuli applied: UPM1-GM in the presence of GM-CSF, UPM1-IL-3 in the presence of IL-3, and UPM1 in the absence of growth factors.

The cell lines were characterized by phenotypic, genotypic, and functional studies. They express myeloid antigens and are strongly positive for LFA-1 (CD18/CD11a). Other markers include CD34, CD38, CD117, HLA-DR, CD13, CD33, CD15, CD71, MPO, CD7, CD69, and CD54. The cell lines also show differences in their growth requirements and response to growth factors, such as IL-3 and GM-CSF. For example, UPM1-GM cells are more responsive to GM-CSF, while UPM1-IL-3 cells are more responsive to IL-3.

The UPM1 cell line was established in the presence of GM-CSF. This is the fastest growing line (doubling time 89 hours), with larger, more...