Hermansky-Pudlak syndrome: clinical presentation and confirmation of the value of the mepacrine-based cytofluorimetry test in the diagnosis of delta granule deficiency

Hermansky-Pudlak syndrome is a rare genetic disease with multiorgan damage secondary to accumulation of ceroid lipofuscin in multiple cytoplasmic organelles. We describe a new family with this syndrome, taking the opportunuty to discuss the role of the mepacrine-based cytofluorimetric test in the evaluation of the related hemorrhagic diathesis.

Hermansky-Pudlak syndrome (HPS) is an autosomal recessive disorder characterized by oculocutaneous albinism and reticuloendothelial deposits of ceroid lipofuscin in macrophages in association with gastrointestinal and pulmonary lesions. The clinical presentation is completed by a moderate bleeding diathesis due to delta storage pool deficiency because of the absence of dense granules.[1,2] The disease is widespread in Puerto Rico where it is usually related to homozygosity for a 16 bp duplication in exon 15 of the HPS gene on chromosome 10q23. There are scanty data on Hermansky-Pudlak in non-Puerto Ricans.[3]

We report three new cases of this syndrome occurring in two sisters and one brother of one Tyrolean family. Except for oculocutaneous albinism, the phenotype of the two sisters differed markedly from that of the brother because of the presence of achondroplasic dwarfism and strabism.

Genetic tests were negative for the 16 bp duplication. Both sisters died at the ages of 57 and 59 years because of severe lung fibrosis. The brother is alive without restrictive lung disease or gastrointestinal colitis. All three patients came to our attention because of spontaneous hemorrhagic diathesis with normal coagulation tests and normal platelet counts.

In addition to a prolonged bleeding time, classical aggregometry showed hypoaggregation with arachidonic acid but normal aggregation with ADP, collagen and ristocetin. Following the methodology described in previous reports,[4] mepacrine-based flow cytometry analysis was performed in all three patients. Briefly, mepacrine, a fluorescent acridine derivative, has a high affinity for adenine nucleotides and accumulates rapidly and selectively into platelet dense granules. When excited at the appropriate wavelength of light, mepacrine emits a green fluorescence.

As shown as example in Figure 1, mean fluorescence was significantly lower in one proband than in a normal control, thus supporting the diagnosis of δ-granule deficiency as definitely confirmed by platelet electron microscopy examination.

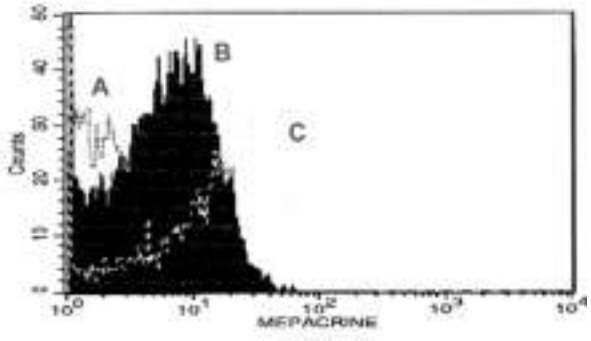

Figure 1. Mepacrine based flow-cytometry histogram. ***A) Blank B) Proband C) Control.***

Several considerations can be drawn from this report: as already suggested by previous papers,[5] the sensitivity of the mepacrine-based cytofluorimetric test in detecting dense granule deficiency is confirmed. However, despite a mild decrease in arachidonic acid-based aggregation, the classic pattern of absent secondary ADP-dependent aggregation waves was not present in our three patients. This conforms with data from Nieuwenhuis *et al.*[6] showing that 33% and up to 35% of patients with delta storage pool disease have a classic aggregation pattern and normal platelet aggregation, respectively. Thanks to its simplicity and reproducibility, cytofluorimetry may therefore be suggested as a complementary test in the routine analysis of suspected storage pool deficiency.[7]

Moreover, these three cases provide evidence of clinical and genetic heterogeneity in Hermansky-Pudlak syndrome in non-Puerto-Rican patients. However, as already observed,[3] and unlike the cases of our patients, the absence of pulmonary fibrosis in non-Puerto Rican subjects is consistently associated with the absence of 16 bp duplication.

In conclusion, we have described three more cases of the rare Hermansky-Pudlak syndrome in the non-Puerto Rican population, reconfirming the role of a simple and reliable method for analyzing dense granule deficiency.

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References

1. Witkop CJ, Townsend D, Bitterman PB, et al. The role of ceroid in lung and gastrointestinal disease in Hermansky-Pudlak syndrome. Adv Exp Med Biol 1989; 266:283-96.
2. Reynolds SP, Davies BH, Gibs AR. Diffuse pulmonary fibrosis and the Hermansky-Pudlak syndrome: clinical course and postmortem findings. Thorax 1994; 49:617-8.
3. Gahl WA, Brantly M, Kaiser-Kupfer MI, et al. Genetic defects and clinical characteristics of patients with a form of oculocutaneous albinism (Hermansky-Pudlak syndrome). N Engl J Med 1998;338:1258-64.
4. Wall JE, Buijs-Wilts M, Arnold JT, et al. A flow cytometric assay using mepacrine for study of uptake and release of platelet dense granule contents. Br J Haematol 1995; 89: 380-5.
5. Gordon N, Thom J, Cole C, et al. Rapid detection of hereditary and acquired platelet storage pool deficiency by flow cytometry. Br J Haematol 1995; 89:117-23.
6. Nieuwenhuis HK, Akkerman JW, Sixma JJ. Patients with a prolonged bleeding time and normal aggregation tests may have storage pool disease: studies on one hundred and six patients. Blood 70;620-3.
7. Schmitz G, Rothe G, Ruf A, et al. European Working Group on Clinical Cell Analysis: Consensus Protocol for the Flow Cytometric Characterisation of Platelet Function. Thromb Haemost 1998; 79:885-96.