Assignment of Human Erythroid $\delta$-Aminolevulinate Synthase (ALAS2) to a Distal Subregion of Band Xp11.21 by PCR Analysis of Somatic Cell Hybrids Containing X:Autosome Translocations

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The erythroid-specific (ALAS2) and housekeeping (ALAS1) genes encoding $\delta$-aminolevulinate synthase have recently been mapped to chromosomes Xp21.1→Xq21 and 3p21, respectively. The erythroid-specific gene is a candidate for mutations resulting in X-linked sideroblastic anemia. Analysis of DNA from hybrid clones containing translocations in the region Xp11.21→Xq21.3 permitted the finer localization of the ALAS2 gene with respect to other loci and breakpoints within this region. These studies localized the ALAS2 gene to the distal subregion of Xp11.21 in Interval 5 indicating the following order: Xpter-OATL2-[L62-3A, Xp11.21; A62-1A-4b, Xp11.21; (ALAS2, DXS323)-(B13-3, Xp11.21; C9-5, Xp11.21;)-(DXS14, DXS429)-DXS422-(DXZ1, Xcen). Thus, the reported linkage of acquired sideroblastic anemia and sideroblastic anemia with ataxia to Xq13 presumably results from genes other than ALAS2.

$\delta$-Aminolevulinate synthase [Succinyl-CoA:glycine C-succinyltransferase (decarboxylating); EC 2.3.1.37, (ALAS)] catalyzes the first committed step in heme biosynthesis. The erythroid-specific gene (ALAS2) has been mapped to the X chromosome and localized to Xp21.1→q21.3 (3). This localization makes ALAS2 a candidate gene for mutations resulting in X-linked sideroblastic anemia (2, 3), consistent with the decreased ALAS activity commonly found in patient’s bone marrow (1). Indeed, the first report of a mutation of ALAS2 in a case of pyridoxine-responsive X-linked sideroblastic anemia (Cotter et al., in press) demonstrates the association between ALAS2 and this disease. However, Raskind et al. (7) reported the linkage of X-linked sideroblastic anemia with ataxia to Xq13, and Dewald et al. (4) found chromosomal rearrangements involving Xq13 in selected patients with idiopathic acquired sideroblastic anemia. In this report, a somatic cell hybrid panel of X:autosome translocations has permitted a finer localization of ALAS2 to the specific subregion Xp11.21.

The somatic cell hybrid panel used to dissect the region Xp21.1→Xq21.3 consisted of the following translocation clones: A2-4, Xp21.1→Xqter; DUA-1CsAzB, Xpter→Xp11.22; F69-3A, Xpter→Xp11.21; A62-1A-4b, Xp11.21; (ALAS2, DXS323)-(B13-3, Xp11.21; C9-5, Xp11.21;)-(DXS14, DXS429)-DXS422-(DXZ1, Xcen). Thus, the reported linkage of acquired sideroblastic anemia and sideroblastic anemia with ataxia to Xq13 presumably results from genes other than ALAS2.

possible to refine the localization of ALAS2 with respect to other loci and breakpoints according to the order: Xpter-OATL2-[L62 3A, Xp11.21]- A62 1A 4b, Xp11.21]- (ALAS2, DXS323)-[A13-3, Xp11.21]- C9-5, Xp11.21]- (DXS11, DXS429)-DXS422-(DXZ1, Xcen). Thus, the ALAS2 locus and DXS323 are subregionally localized to the distal segment of Xp11.21, as defined by the incontinentia pigmenti-1 (IP-1) breakpoints within hybrid clones B13-3 and C9-5. The proximal segment of Xp11.21 presumably contains the IP-1 locus and additional loci as noted in Gorski et al. (5). The ordering of these loci relative to each other within Xp11.21 awaits further study. This mapping of ALAS2 to the distal subregion of Xp11.21 also suggests that genes other than or in addition to ALAS2 are involved in the X-linked and idiopathic-acquired sideroblastic anemias associated with Xq13.

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REFERENCES