The effect of putative nucleation sites on the loading and stability of iron in ferritin

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摘要

Abstract

The L chain of the iron storage protein ferritin contains more putative nucleation sites in the core (Glu53, 56, 57, 60, and 63) than does the H chain (Glu61, 64, and 67). Recombinant DNA techniques were used to investigate the role of these putativenucleation sites on iron loading by ceruloplasmin and on the stability of the iron core. Recombinant rat liver ferritin H chain homopolymer and the two mutants (E61A and EB1A-E64A), containing three, two and one nucleation sites, respectively, loaded up to 2010 +/- 50, 2010 +/- 40, and 1950 +/- 40 atoms of iron per ferritin, respectively. However, the mutations resulted in a 50% decrease in the rate of iron loading by ceruloplasmin. The ferritin variants incorporated the same amount of phosphate after iron loading (410 +/- 20, 400 +/- 30, and 420 +/- 20 atoms per ferritin, respectively). The stability of the iron cores prior to phosphate incorporation, assessed by the rate of iron release by 10 mM EDTA and the paraquat cation radical, corresponded to numbers of proposed nucleation sites. The subsequent incorporation of phosphate seemed to stabilize the iron core and minimized the effect of numbers of putative nucleation sites in ferritin on the rate of iron release by EDTA and the paraquat cation radical. After incorporation of phosphate the ferritins behaved similarly to the native rat liver ferritin with respect to the rate of iron release by the paraquat cation radical. (C) 1998 Academic Press. [References: 32]