Auctoclavable archaeal ferritin

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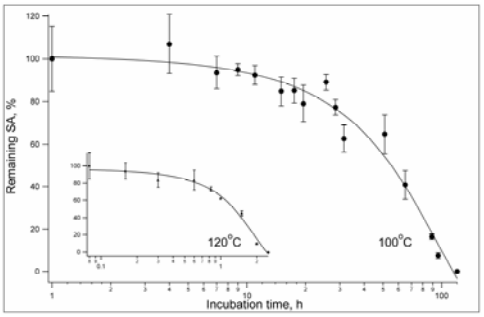
A ferritin from the obligate anaerobe and hyperthermophilic archaeon *Pyrococcus furiosus* (optimal growth at 100 ^{o}C) has been cloned and overproduced in *Escherichia coli* to one-fourth of total cell free extract protein, and has been purified in one step to homogeneity by incubating at 100 ^{o}C for 30 minutes.

The produced archaeal ferritin (PfFtn) is structurally similar to the known bacterial and eucaryal ferritins; it is a 24-mer of 20 kDa subunits, which add up to a total Mr 480 kDa. The isoelectric point is 4.5. The protein belongs to the non-heme type of ferritins. The 24-mer contains approximately 17 Fe (as isolated), 2700 Fe (fully loaded), or <1 Fe (apoprotein). Fe-loaded protein exhibits an EPR spectrum characteristic for superparamagnetic core formation. At 25 ^{o}C Vmax = 25 µmole core Fe(III) formed per min per mg protein when measured at 315 nm, and the $K_{0.5}$ = 5 mM Fe(II). At 0.3 mM Fe(II) activity increases 100-fold from 25 to 85 ^{o}C.

The protein is extremely thermostable; its ferrihydride core-formation activity has a half-life of 48 hours at 100 ^{o}C and 85 minutes at 120 ^{o}C. The extreme thermostability of PfFtn has potential value for biotechnological applications.



PfFtn crystals have been produced and a preliminary crystal structure has been determined[1].

1. Matias P.M., Tatur J., Carrondo M.A., Hagen W.R. Acta Cryst. Sect. F *in press*.