

Supplementary Information

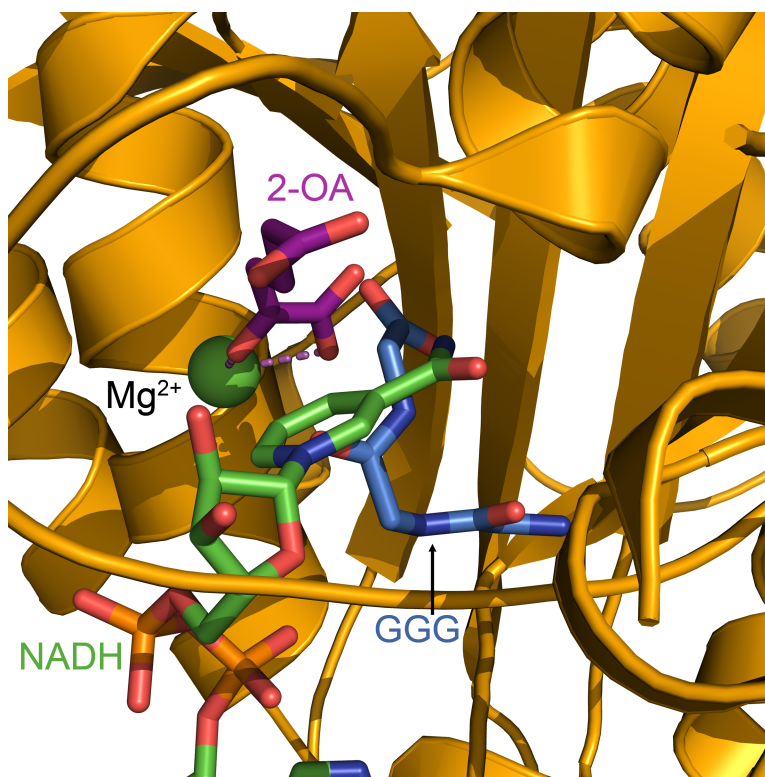


Figure S1. Comparison of the orientation of the GGG tripeptide bound to SpHICDH with the modeled products 2-OA and NADH. Ribbon diagram of SpHICDH (orange) bound to GGG (blue) overlaid with the products 2-OA (violet), NADH (green) and Mg²⁺ (green sphere). The products were modeled by aligning SpHICH-GGG with ScICDH bound to NADPH and 2-OG (PDB code 2QFX). The tripeptide binds in the active site and occupies regions of the 2-OA and NADH binding pockets.

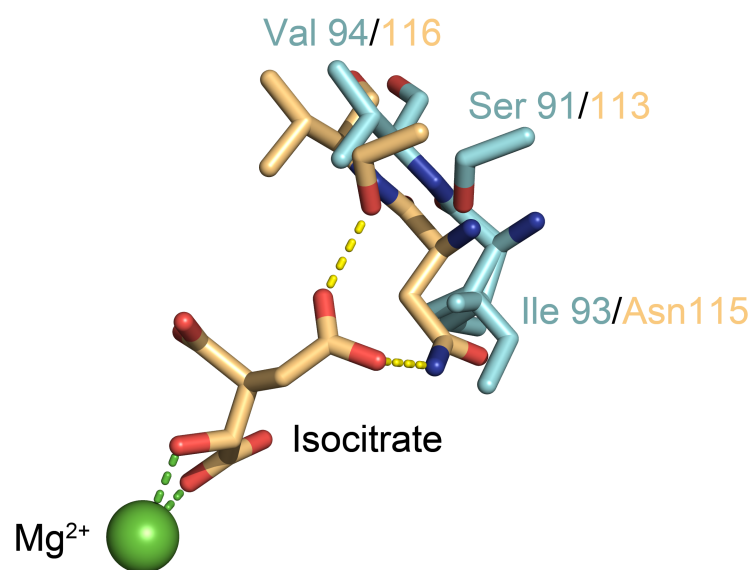


Figure S2. Structural alignment of key residues that determine substrate specificity in EcICDH and SpHICDH. SpHICDH apoenzyme (cyan) was overlaid with EcICDH in a complex with isocitrate and NAD⁺ (PDB code 1AI2, light orange). Residues that confer binding specificity of EcICDH to isocitrate are labeled in light orange, with the corresponding residues in SpHICDH labeled in cyan.