



**Supplemental Data Figure 4.** Structural analysis of cofactor binding to 17βHSD1. Panel A (cyan) represents the apo-enzyme; Panel B (green) NAD<sup>+</sup> bound form; Panel C (salmon) NADP<sup>+</sup> bound form; Panels D, superimposition of all three structures; and Panel E, superimposition of the bound cofactors, extracted from these structures. These images and the discussion above demonstrate that cofactor binding to HSDs and related SDR enzymes and cofactor selectivity or preference is influenced by:

1. Structural elements, like helix-loop transitions, loop stabilization by the ligand(s), length of loops and helices in the Rosman Fold;
2. Variability in the primary sequence for an equivalent position in the structure;
3. Conformational changes in the side chain of identical amino acids for a specific position in the structure, and the influence of structural elements in such changes.