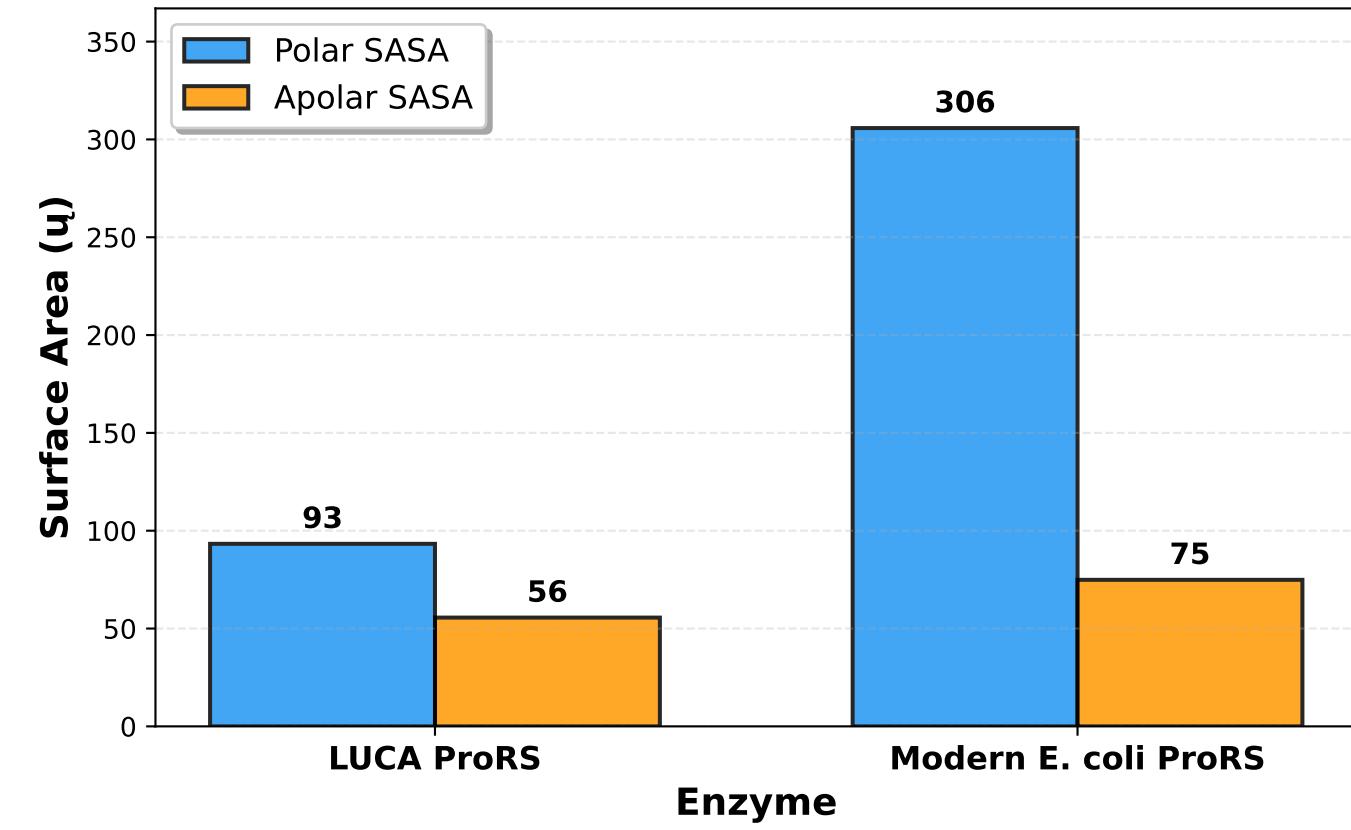
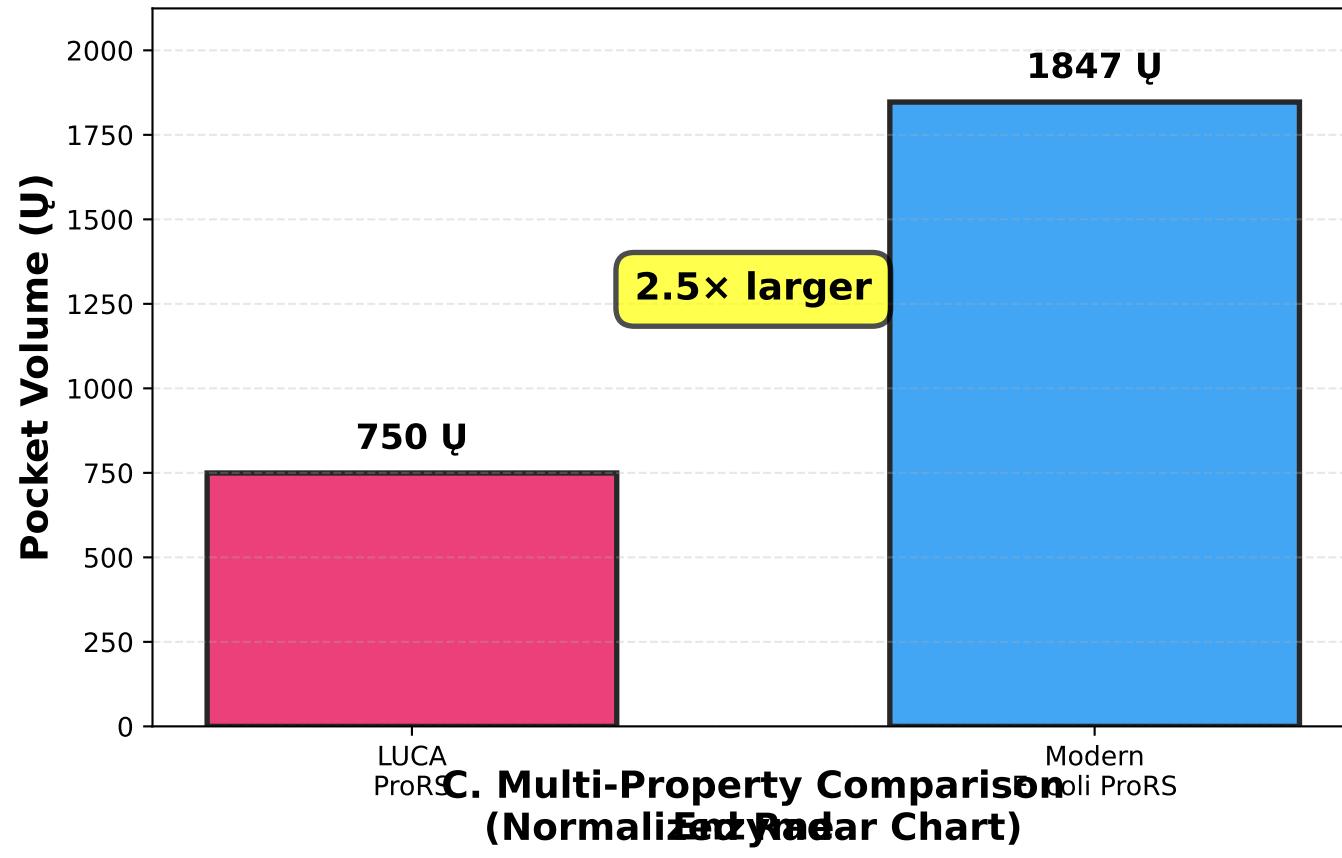


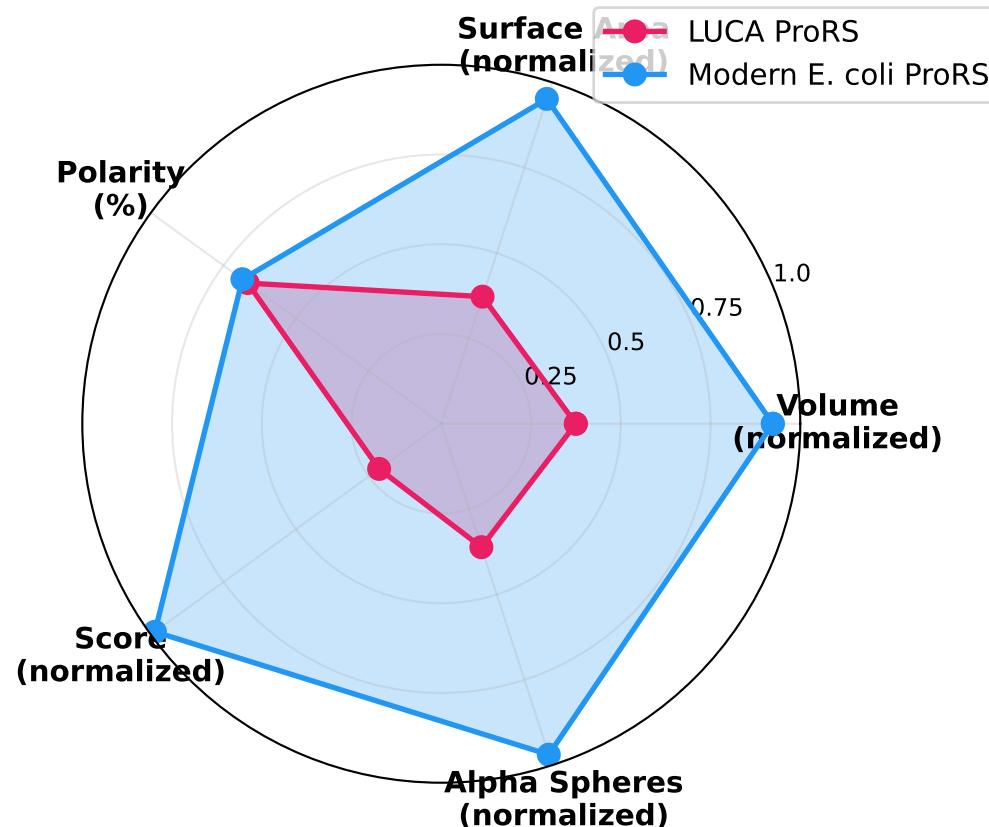
Figure 6: Prokaryotic Binding Pocket Analysis - Evolution from LUCA to Modern

Modern is 2.5x Larger than LUCA



D. Summary Table: Modern Pocket is Larger & Higher Quality

Property	LUCA ProRS	Modern E. coli	Fold Change
Volume (\AA^3)	750	1847	2.5x
Total SASA (\AA)	149	381	2.6x
Polar SASA (\AA)	93	306	3.3x
Polarity (%)	66.7	68.5	~Equal
fPocket Score	0.32	1.48	4.6x
Alpha Spheres	47	126	2.7x



KEY FINDINGS:

1. VOLUME EXPANSION: Modern pocket is 2.5x larger (1847 vs 750 \AA^3)
 - More spacious substrate binding site
 - Potentially accommodates diverse conformations
2. SURFACE AREA: Modern has 2.6x more surface area (381 vs 149 \AA)
 - Increased protein-ligand interactions
 - More contact points for substrate recognition
3. POLARITY MAINTAINED: Both ~67-68% polar
 - Hydrophilic character conserved across evolution
 - Consistent with binding charged amino acids (Pro, Thr)
4. QUALITY SCORE: Modern scores 4.6x higher (1.48 vs 0.32)
 - Better-defined binding pocket geometry
 - More optimal for substrate binding
5. COMPLEXITY: Modern has 2.7x more alpha spheres (126 vs 47)
 - More sophisticated pocket architecture
 - Refined geometric organization

BIOLOGICAL INTERPRETATION:
Evolution EXPANDED the binding pocket while maintaining its chemical properties (polarity). Larger volume may allow promiscuous binding of both Pro and Thr, consistent with ipTM data (Figure 3).