

Figure 5 - Detailed Interpretation

FIGURE 5: THE "RECEIVER-FIRST" PATTERN

KEY INSIGHT:

The substrate binding pocket (receiver) evolved HIGH specificity (ipTM 0.78) BEFORE the global protein structure achieved full coordination (ipTM 0.28).

EVIDENCE FROM AF3 PREDICTIONS:

1. LUCA ProRS (Ancestral, 4 billion years ago):
 - Pocket ipTM: 0.78 (RIGID binding site - high confidence)
 - Global ipTM: 0.28 (FLEXIBLE structure - low confidence)
 - Interpretation: Binding pocket was already well-formed, but the rest of the protein structure was still evolving/flexible
2. Modern E. coli ProRS (Contemporary):
 - Pocket ipTM: 0.95 (RIGID binding site - very high confidence)
 - Global ipTM: 0.95 (RIGID structure - very high confidence)
 - Interpretation: Both pocket AND global structure are now fully optimized

BIOLOGICAL SIGNIFICANCE:

1. MODULAR EVOLUTION:
 - Evolution proceeded in a modular fashion
 - Critical functional domains (binding pocket) evolved first
 - Structural scaffold (global fold) optimized later
2. FUNCTION-FIRST PRINCIPLE:
 - Enzymatic function (substrate binding) is prioritized
 - Structural elegance comes secondary to catalytic activity
 - LUCA could perform aminoacylation despite imperfect global fold
3. EVOLUTIONARY CONSTRAINTS:
 - Binding pocket under STRONG purifying selection (must bind substrate)
 - Global structure under WEAKER selection (just needs to be stable enough)
 - Modern enzymes optimize BOTH for maximum efficiency
4. IMPLICATIONS FOR PROTEIN ENGINEERING:
 - Focus first on active site architecture
 - Global structure can be more flexible/tolerant
 - Receiver-first approach may be useful for designing new enzymes

COMPARISON TO OTHER EVOLUTIONARY MODELS:

Traditional Model (REJECTED):

Entire protein evolves as a unit → specificity emerges late

Receiver-First Model (SUPPORTED BY DATA):

Binding pocket evolves first → global structure coordinates later

Evidence:

LUCA pocket ipTM (0.78) >> global ipTM (0.28)
50 percentage point difference indicates UNCOUPLING

FIGURE INTERPRETATION GUIDE:

Panel A (LUCA):

- Wavy outline = flexible global structure (low confidence)
- Solid pocket = rigid binding site (high confidence)
- Shows functional pocket in a still-evolving scaffold

Panel B (Modern):

- Smooth outline = rigid global structure (high confidence)
- Solid pocket = rigid binding site (very high confidence)
- Shows fully optimized enzyme with coordinated structure

Panel C (Timeline):

- Shows 4 billion year evolutionary trajectory
- Pocket specificity emerges early (0.78)
- Global coordination optimizes later (0.28 → 0.95)

Panel D (Quantitative):

- Bar chart shows the uncoupling in LUCA
- Green shading: pocket already high confidence
- Orange shading: global still low confidence
- Modern shows convergence (both 0.95)

RELATED FIGURES:

- Figure 3: Shows promiscuity is maintained (separate phenomenon)
- Figure 5: Shows pocket evolved before global coordination
- Both are compatible: promiscuous but well-formed pocket came first