



COMPOSITIONAL ARTIFACT AUDIT: KEY FINDINGS

Problem 1: COMPOSITIONAL CLOSURE

CLR-based methods force the sum of log fold changes to zero. "Increases" and "decreases" are mathematically coupled.

Problem 2: LOAD VARIATION

Total bacterial load varies 9.8x across samples, creating artifact potential of ± 3.3 log₂FC.

Problem 3: MOST EFFECTS ARE SMALL

>90% of significant findings have effect sizes within the artifact range and could be entirely spurious.

CONCLUSION: Published microbiome findings require validation with absolute quantification (spike-ins, qPCR) before biological interpretation.