

## Conclusion

M13's high predicted toxicity results from coupling/dimerization creating a rigid 3-ring structure (C21H14Br6O3) with enhanced lipophilicity (XLogP=6.97, Z=+2.15) and retained phenolic OH groups that drives mitochondrial membrane potential disruption (SR-MMP: 99.6%) and oxidative stress response activation (SR-ARE: 81.0%).

## Methods

I analyzed M13 using a systematic 6-step approach: (1) Data isolation from merged toxicity/descriptor datasets using pandas, (2) Structural characterization using RDKit to count bromines, phenolic OH groups, and aromatic rings relative to TBBPA, (3) Statistical anomaly detection by calculating Z-scores for all 51 molecular descriptors and ranking by absolute magnitude, (4) Risk endpoint identification by filtering for HIGH risk levels and extracting corresponding probabilities, (5) Causal chain synthesis linking transformation→structure→descriptors→endpoints→mechanism, (6) Validation plan development with specific experimental protocols. RDKit was used for SMILES parsing and molecular property calculation. Statistical analysis used numpy for Z-score calculations [(value - mean) / std] across the 21-molecule cohort.

## Results

M13 structural analysis: Coupling/dimerization transformation created a 3-ring structure (vs 2 for TBBPA) with molecular formula C21H14Br6O3, MW 793.76 g/mol (1.46× TBBPA), 6 bromines (+2 vs TBBPA), and 2 retained free phenolic OH groups. Top 5 anomalous descriptors (Z-scores): Polar\_Area 40.70 (Z=-2.37), Nonpolar\_Area 59.32 (Z=+2.35), XLogP 6.97 (Z=+2.15), Neg\_Average -0.0161 (Z=+1.81), Pi 0.0171 (Z=-1.78). Four HIGH risk endpoints: SR-MMP 0.9960, Eye\_irritation 0.9210, Respiratory\_toxicity 0.9021, SR-ARE 0.8102. M13 showed the highest mitochondrial toxicity probability (99.6%) in the entire 21-molecule dataset.

## Challenges

The small dataset size (n=21) limited statistical power for more sophisticated multivariate analyses. LEA\_Var mentioned in the hypothesis was found but showed only moderate anomaly (Z=+0.59), not ranking in top 5. The hypothesis prediction of "high LEA\_Var" was not supported by the data. Some risk level classifications used "HIGH" rather than "High" requiring case-sensitive string matching. The 3D visualization of molecular structures was not implemented due to focus on quantitative descriptor analysis. RDKit structure counting required careful SMARTS pattern selection for accurate phenolic OH group identification.

## Discussion

The analysis confirms the research hypothesis with modifications. M13's coupling transformation creates a unique "toxic triad": enhanced lipophilicity (XLogP 2.15 SD above cohort mean), retained redox-active phenolic groups (2 free OHs), and rigid 3-ring membrane-inserting structure. This combination drives both mitochondrial membrane disruption (SR-MMP: 99.6%) and oxidative stress response (SR-ARE: 81.0%), consistent with a dual mechanism of lipophilic membrane accumulation and phenolic redox cycling. The anomalous descriptor pattern (↓Polar\_Area, ↑Nonpolar\_Area, ↑XLogP) aligns with SHAP model drivers mentioned in the manuscript, supporting the prediction that structural coupling creates compounds with enhanced membrane partitioning potential while maintaining reactive phenolic functionality.

## Proposed Next Hypotheses

The phenolic OH count × XLogP interaction term is the primary driver of SR-MMP toxicity across

all coupling/dimerization products (M08-M13), with compounds having  $\geq 2$  phenolic groups and XLogP  $>6.0$  showing SR-MMP probabilities  $>0.9$ . M13's unique ether linkage configuration stabilizes mitochondrial membrane insertion compared to other coupling products, explaining its highest SR-MMP probability (99.6%) within the coupling class despite similar OH counts.

