Conclusion

The high predicted toxicity of molecule M12 stems from incomplete methylation that retains one phenolic OH toxicophore while exhibiting anomalous electronic properties (lower nucleophilicity, electrostatic potential, and electronic variance), leading to HIGH-risk predictions for mitochondrial (SR-MMP), respiratory, eye irritation, and CYP1A2 inhibition endpoints.

Methods

Analyzed M12 data by: (1) merging toxicity predictions, molecular descriptors, and transformation classes datasets on SMILES identifiers; (2) extracting M12-specific data and characterizing its structure using RDKit SMARTS patterns for phenolic OH ([OH]c), methoxy (cOC), and bromine groups ([Br]c); (3) calculating Z-scores for all 51 molecular descriptors relative to the 21-molecule cohort mean and standard deviation; (4) identifying HIGH-risk toxicity endpoints and their probabilities; (5) comparing M12 to other molecules with similar structural features; (6) synthesizing findings into a mechanistic causal chain linking transformation → structure → descriptors → toxicity endpoints.

Results

M12 (C16H14Br4O2, MW=557.90) contains 1 free phenolic OH, 1 methoxy group, and 4 aromatic bromines, representing incomplete methylation. The 5 most anomalous descriptors (Z-scores): Nu (-1.88), Overall_Variance (-1.82), ESPmax (-1.69), Pos_Average (-1.61), and Pi (-1.60). M12 exhibits HIGH risk for 4 endpoints: SR-MMP (p=0.944), Respiratory toxicity (p=0.909), Eye irritation (p=0.905), and CYP1A2 inhibition (p=0.887). Comparison with other 1-OH molecules shows M12 has among the highest probabilities for SR-MMP and CYP1A2 endpoints.

Challenges

The primary analytical challenge was the absence of parent TBBPA in the dataset, limiting direct before/after methylation comparisons. The small sample size (n=21) restricted statistical power for subgroup analyses. Additionally, M12 is the sole methylation product, preventing within-transformation-class comparisons to assess methylation effects systematically.

Discussion

M12 represents a bioactivation case where incomplete methylation fails to mask the phenolic toxicophore while altering electronic properties. The retained OH group enables quinone formation and redox cycling, explaining the HIGH mitochondrial toxicity (SR-MMP) prediction. Lower nucleophilicity and electrostatic potential suggest altered reactivity that may enhance membrane permeability and cellular target interactions. This contradicts the traditional assumption that methylation is always detoxifying, demonstrating that incomplete conjugation can maintain or enhance toxicity.

Proposed Next Hypotheses

Incomplete methylation of other polyphenolic compounds (e.g., catechols, hydroquinones) will similarly fail to reduce toxicity when one or more OH groups remain unmethylated. The degree of toxicity reduction from methylation depends on the ratio of methylated to free OH groups, with complete methylation required for effective detoxification.

Artifacts

Artifact 1:

File name: M12_anomalous_descriptors_and_toxicity.png

Artifact description: A two-panel figure summarizing M12's anomalous molecular properties. Panel A shows the top 5 molecular descriptors with the largest Z-score deviations from the cohort

(all negative, indicating lower values). Panel B displays M12's 4 HIGH-risk toxicity endpoints with their predicted probabilities, all exceeding 85% threshold. Created using matplotlib with horizontal bar charts for clear visualization of the bioactivation mechanism evidence.