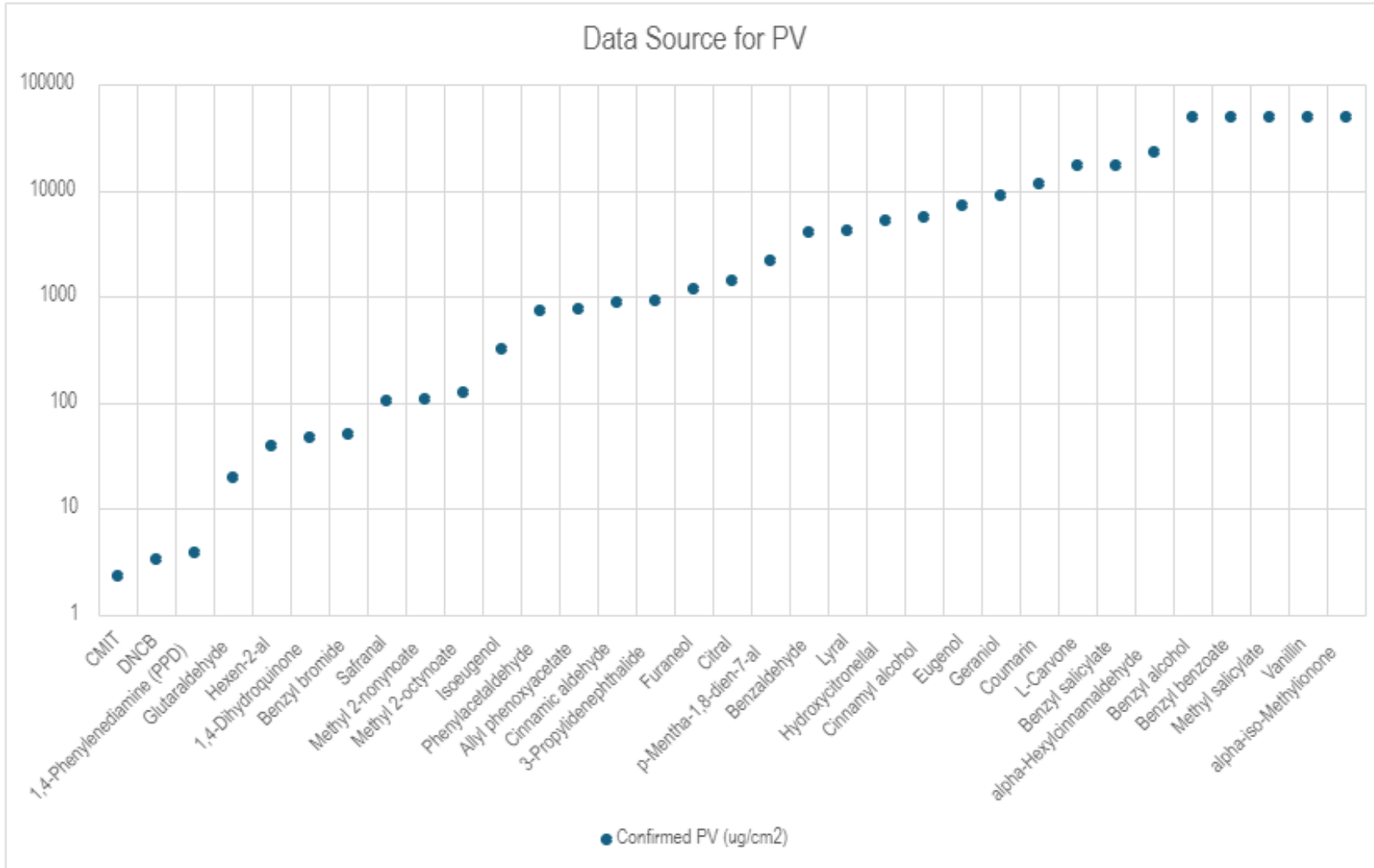


**In vitro data and prediction approaches:
initial results comparing the RCPL with
data from the GARD-DR and Linear
Regression models**

P. Griem

RCPL33

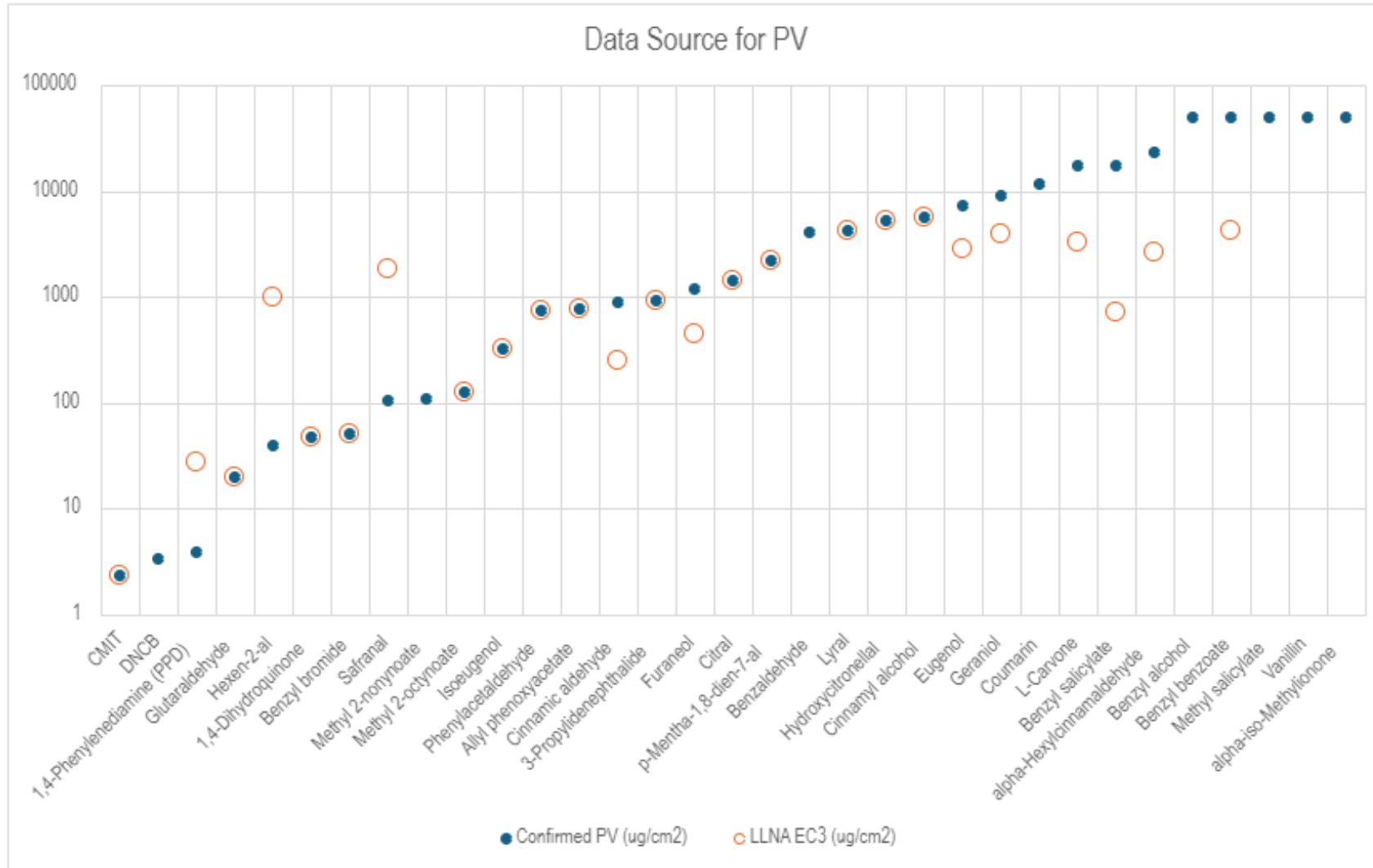
PVs span 4 orders of magnitude



PV span 4 orders of magnitude

RCPL33 – Derivation Basis for PVs

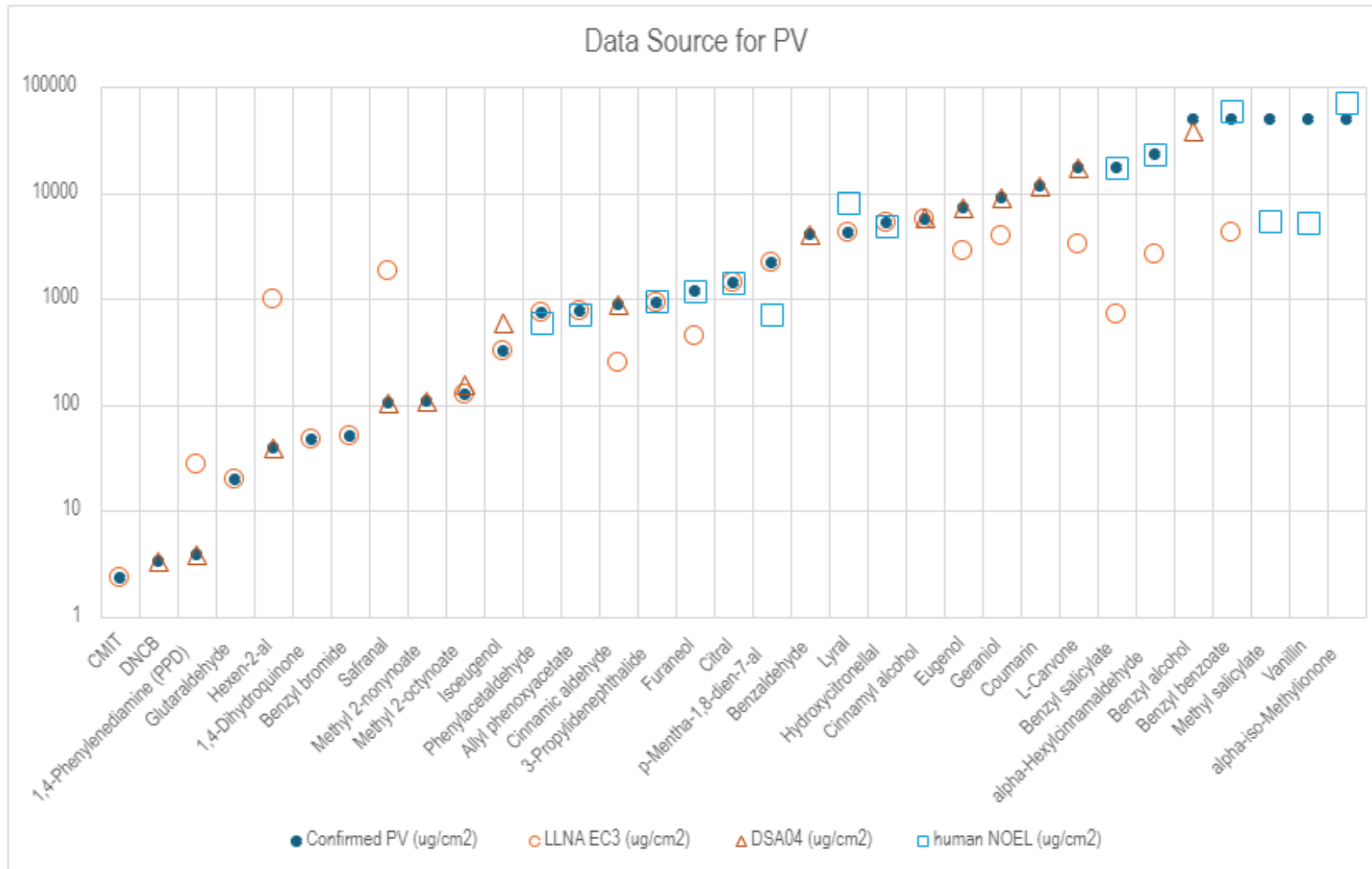
LLNA EC3



- LLNA EC3 values displayed
- PV circled are based on EC3

RCPL33 – Derivation Basis for PVs

LLNA EC3 – DSA04 – human HRIPT NOEL



- Human DSA04 (where available) or human NOEL values displayed
- PV framed by triangle or square are based on human data

Linear Regression model EQ5

Method



EQ5 Method

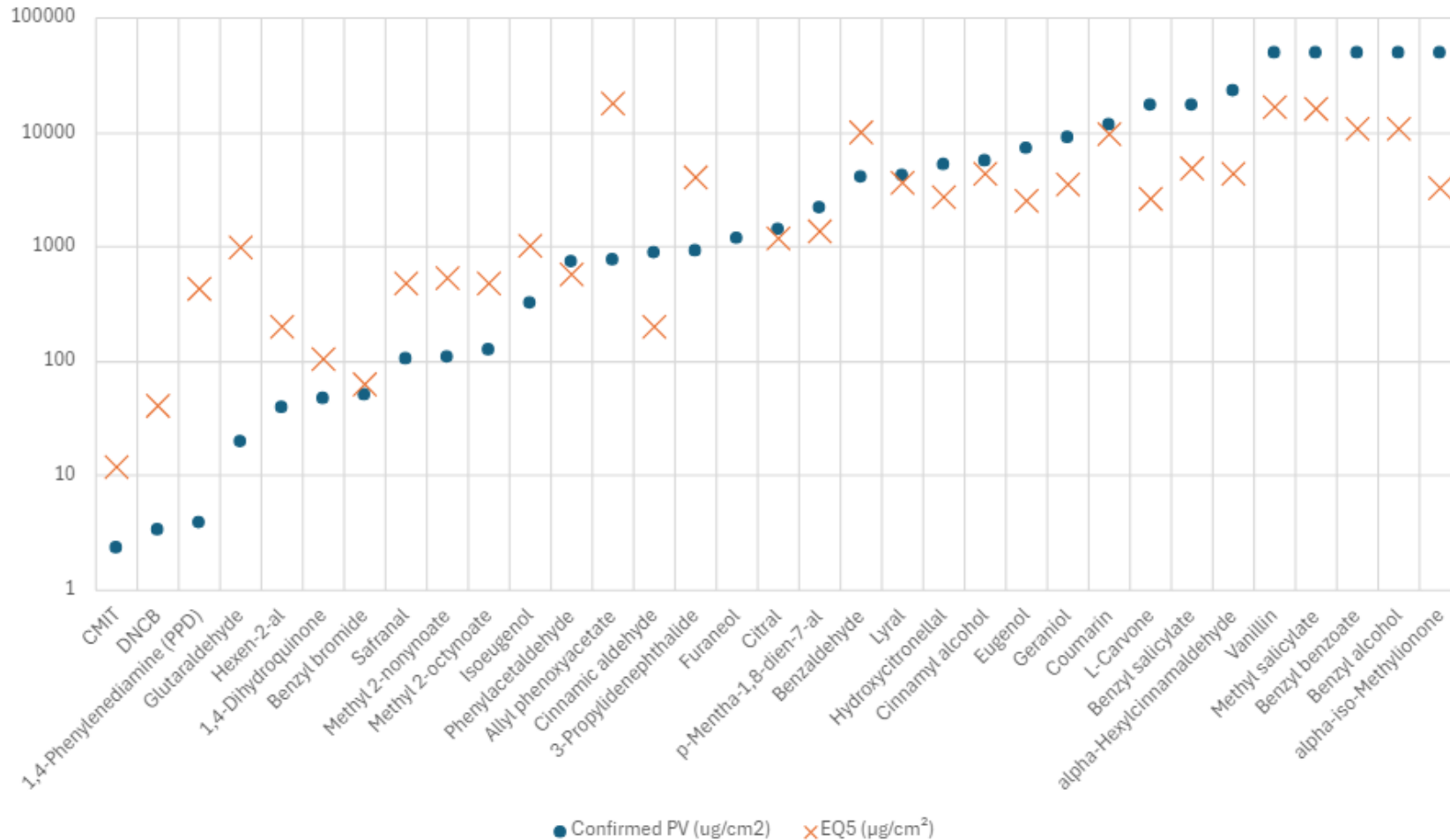
- Input parameters:
 - kDPRA maximum normalized logarithmic rate constant log Kmax norm
 - KeratinoSens normalized IC50 (concentration reducing cell viability by 50%)
 - KeratinoSens normalized EC1.5 (concentration causing 1.5fold induction of luciferase activity)
 - h-CLAT normalized MITnorm as lowest concentration leading to 1.5fold CD86 or 2fold CD54 expression induction
 - h-CLAT CV75norm normalized concentration reducing viability by 25%
 - VPnorm normalized vapor pressure describing volatility from LLNA vehicle
- Trained on LLNA EC3 values
- $$pEC3 = 0.20 + 0.34 \times \text{Log } k_{\text{max norm}} + 0.20 \times \text{Log } \text{MITnorm} + 0.09 \times \text{Log } \text{EC1.5norm} + 0.21 \times \text{Log } \text{CV75norm} + 0.11 \times \text{Log } \text{IC50norm} - 0.19 \times \text{Log } \text{Vpnorm}$$

RCPL33 – PVs vs. Predictions

Linear regression EQ5



PV vs Prediction



Linear regression EQ5 values based on in vitro sensitization battery (kDPRA, KS, hCLAT) shows good correlation with derived potency values

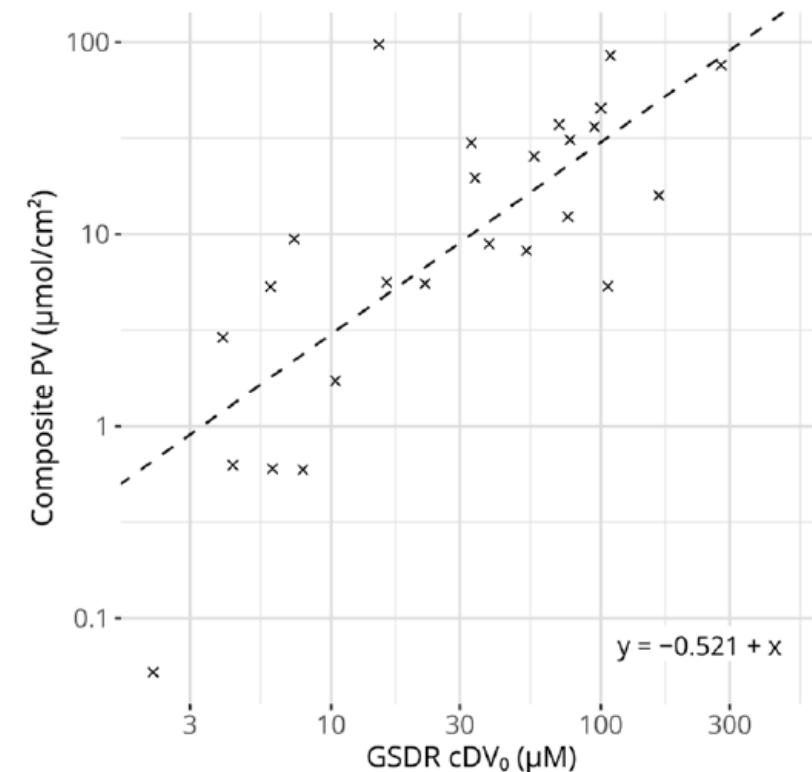
GARDskin DoseResponse

Genomic Allergen Rapid Detection - Method



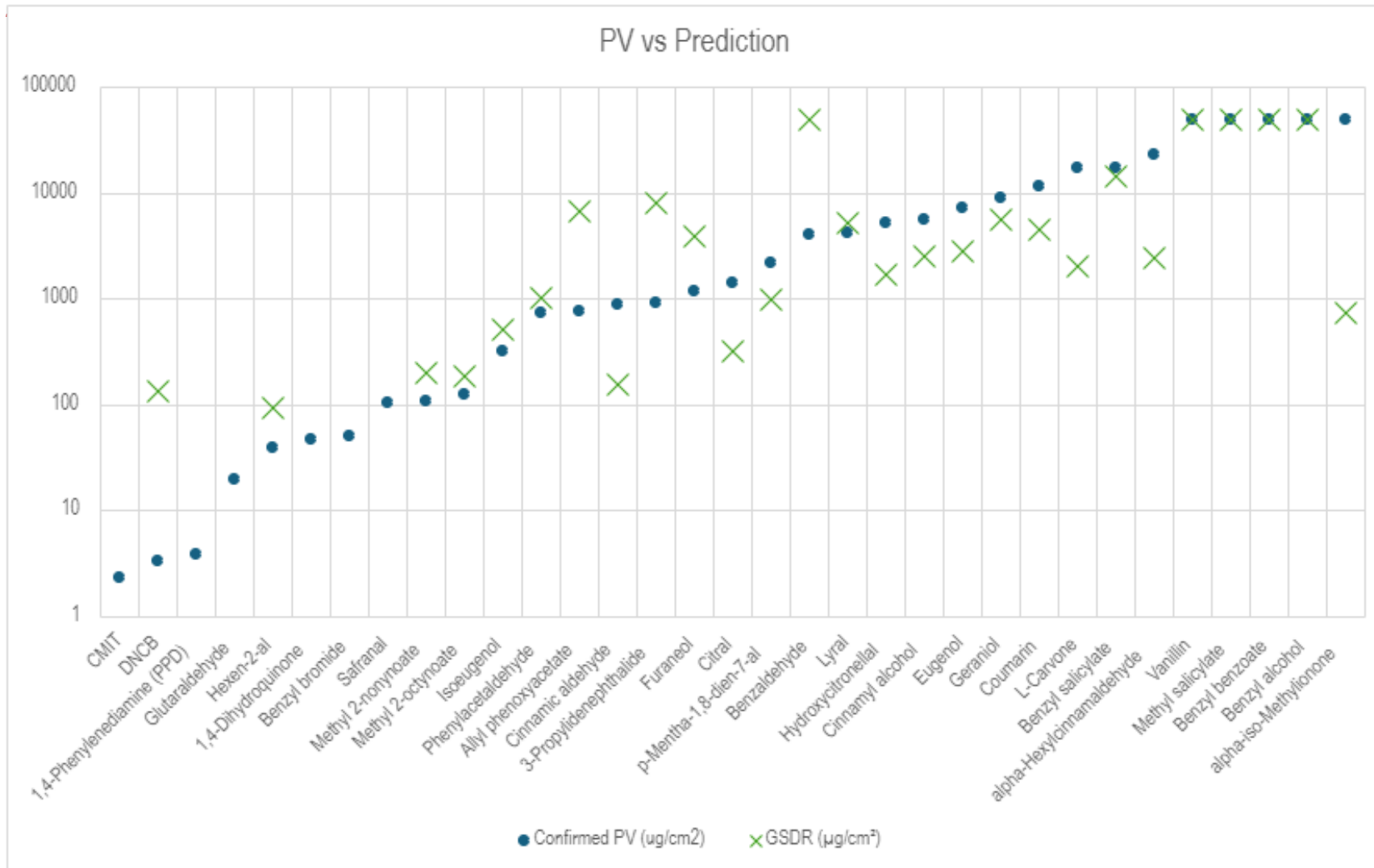
GARD DR method:

- SenzaCells (hu DC-like) incubated with different concentrations of test chemical for 24 hours
- Total RNA is isolated and purified
- Gene expression analysis of selected marker gene set (eg, xenobiotic recognition, antigen presentation and co-stimulation, and induction of cellular and oxidative stress pathways)
- cDV0 as interpolated lowest concentration at which response is observed
- CDV0 relationship to LLNA EC3 and human HRIPT NOELs (combined into composite potency value)
- Predicted potency in $\mu\text{mol}/\text{cm}^2 = \text{cDV0 in } \mu\text{M} \times 0.301$
- Predicted potency in $\mu\text{g}/\text{cm}^2 = \text{cDV0 in } \mu\text{g}/\text{mL} \times 304$



RCPL33 – PVs vs. Predictions

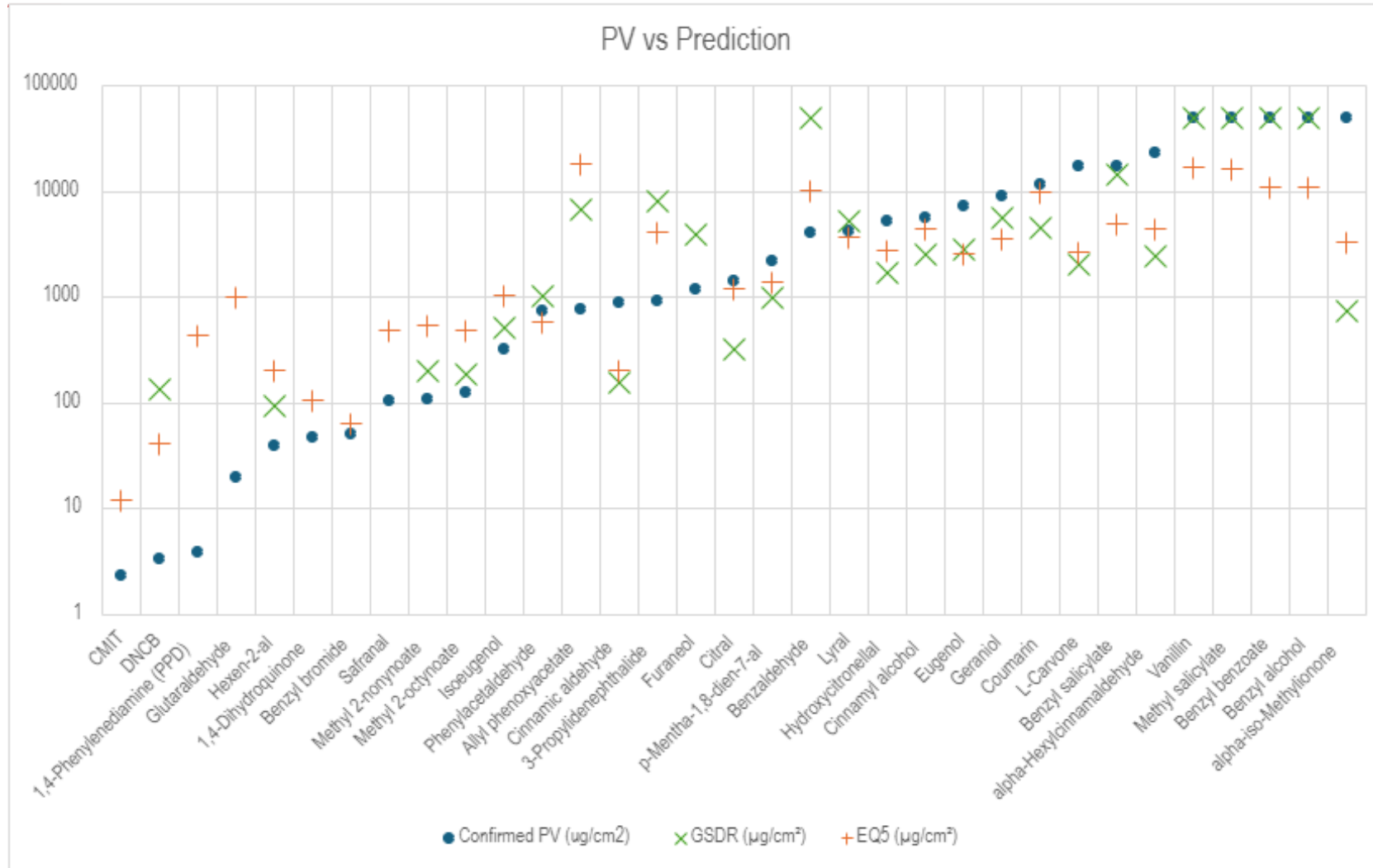
GardSkin DoseResponse (GSDR)



GARDskin DoseResponse values based on in vitro gene expression analysis shows good correlation with derived potency values

RCPL33 – PVs vs. Predictions

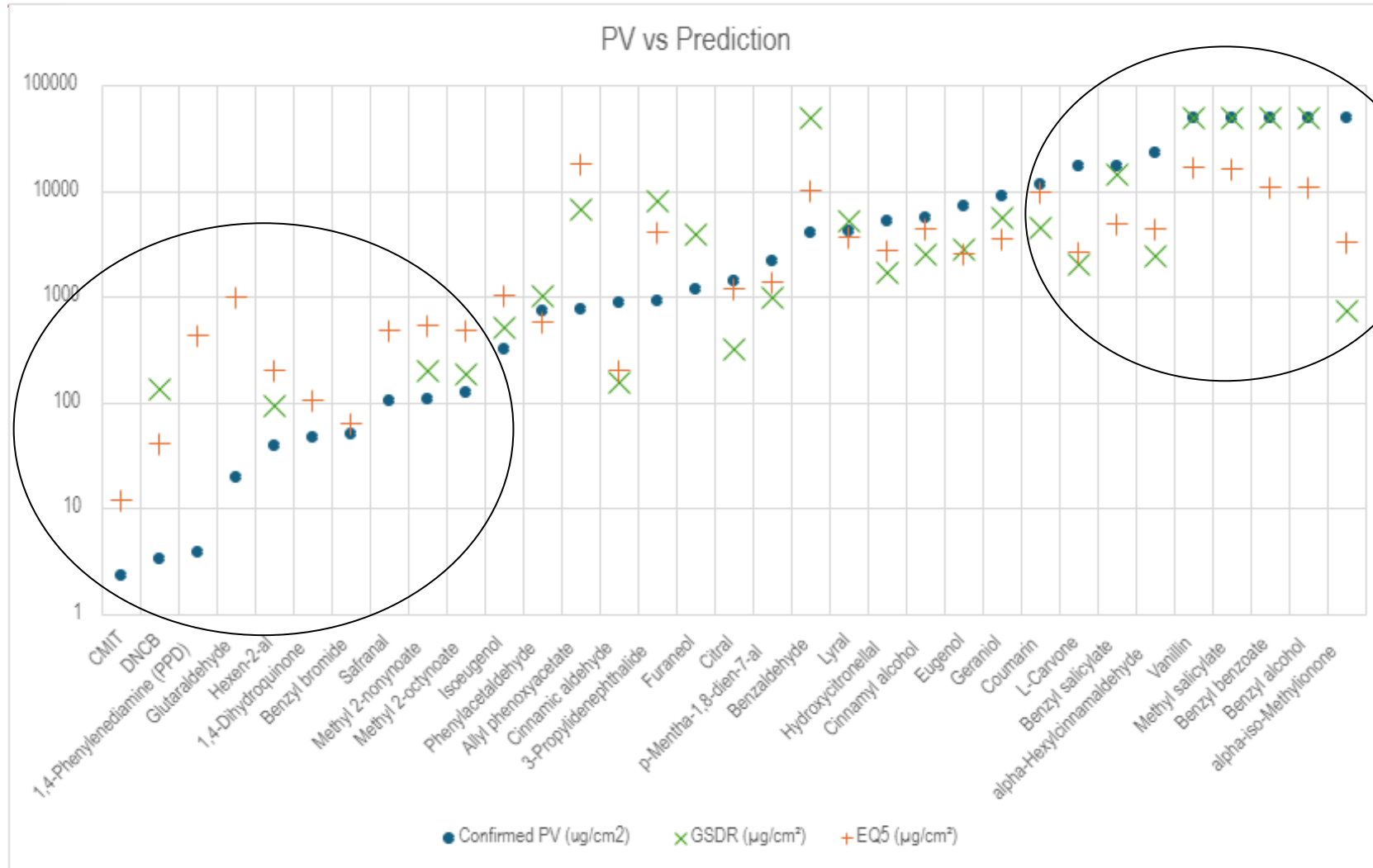
EQ5 and GSDR



- Overall, no obvious difference in prediction quality between EQ5 and GSDR
- GSDR concluded 'non-sensitizer' for 4/5 chemicals that were considered 'very weak to non-sensitizing' during potency evaluation
- Both predictions consistently fall on either the higher or lower side of the derived PV
- This is no surprise as GARD analysis genes which are also involved in the cellular responses measured in the set of in vitro tests

RCPL33 – PVs vs. Predictions

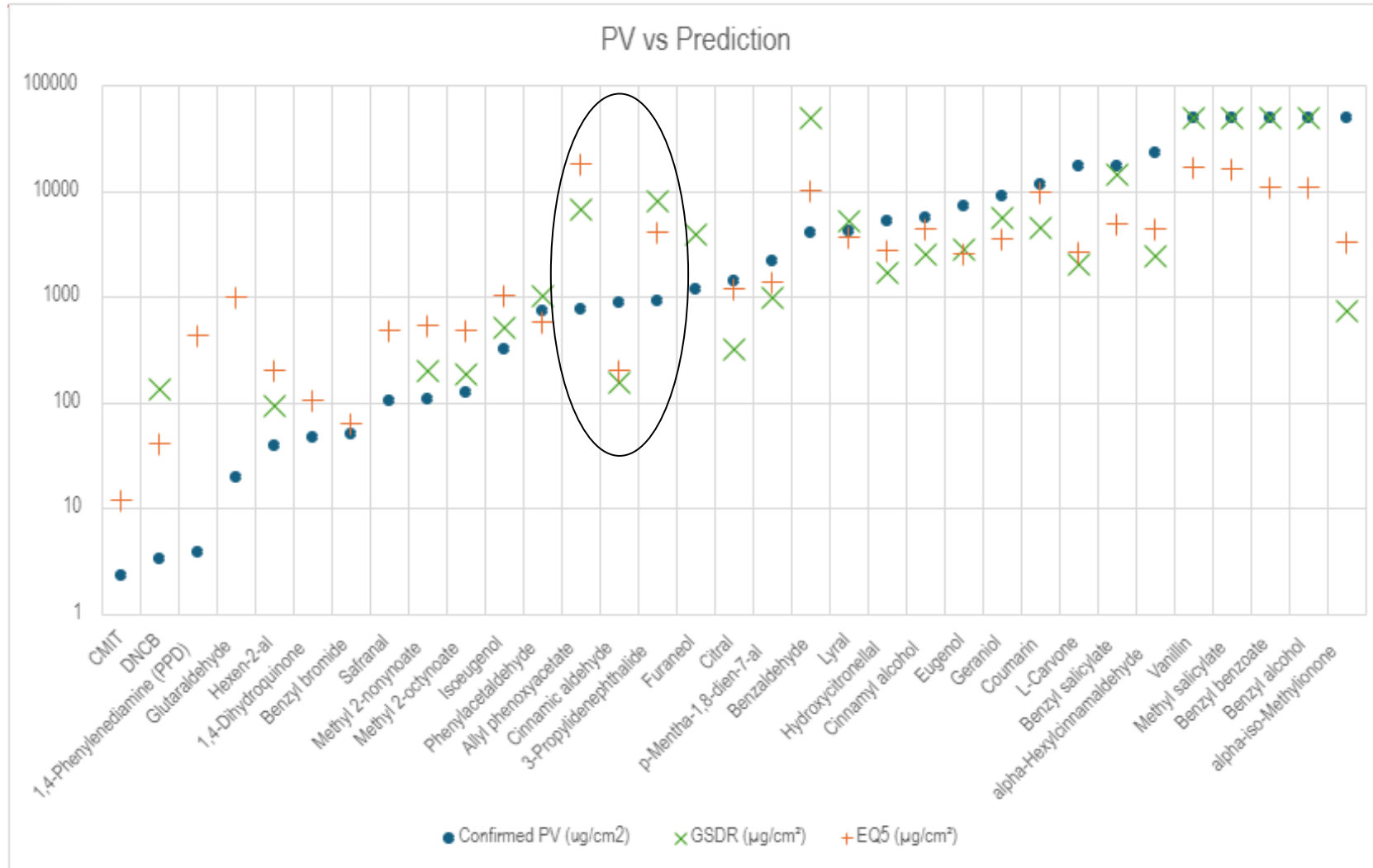
EQ5 and GSDR – vs. LLNA EC3



- NAM-based prediction models tend to
- Underestimate potency for very strong sensitizers
 - Overestimate potency for very weak sensitizers

RCPL33 – PVs vs. Predictions

EQ5 and GSDR – vs. LLNA EC3



Even for moderate potency sensitizers NAM-based predictions can vary considerable from PV value depending on the chemical

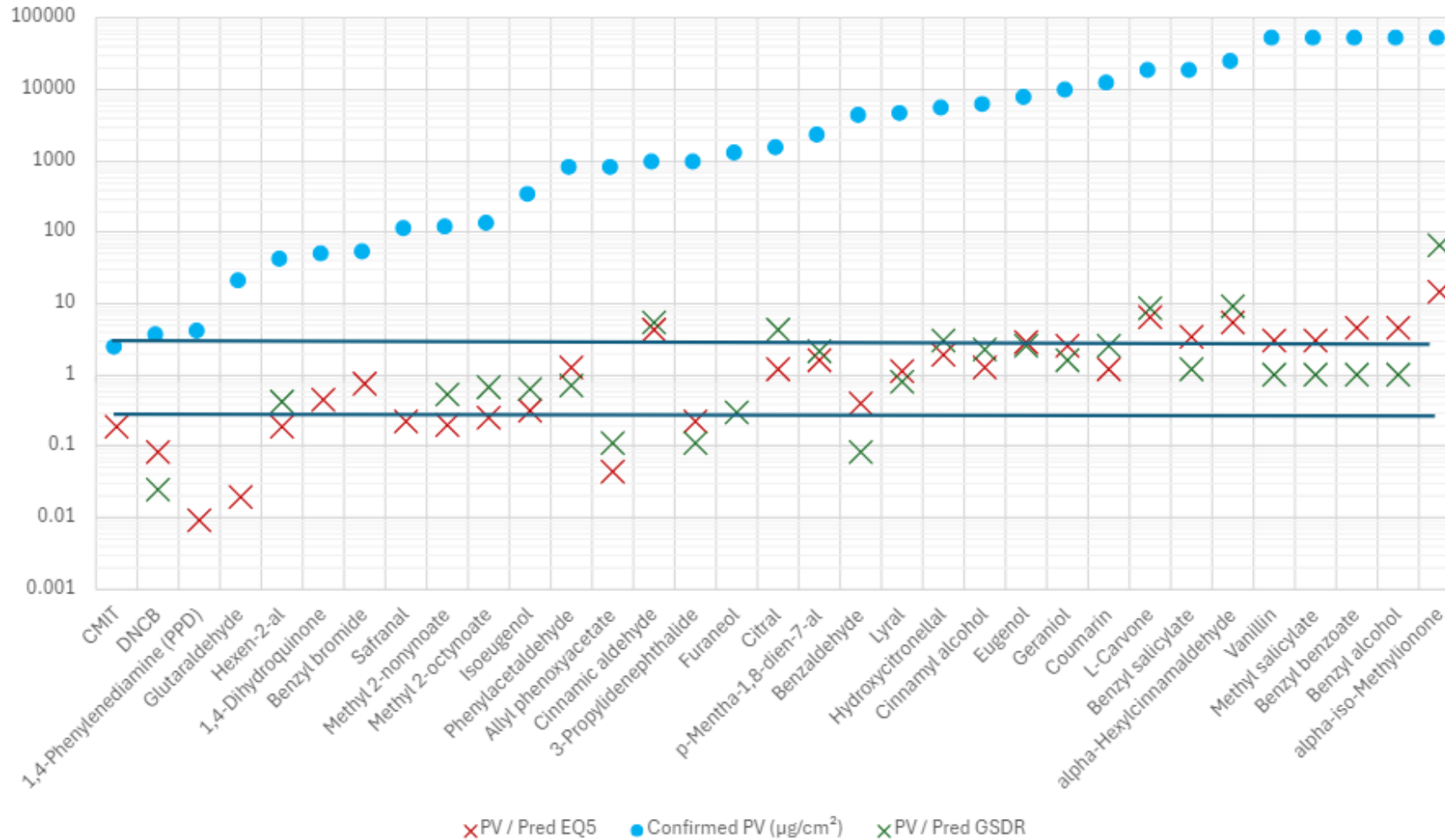
- Is this caused by one or more identifiable parameters?

RCPL33 – PVs vs. Predictions

Ratio of PV and mean of EQ5 and GSDR



PV / Pred Mean Analysis



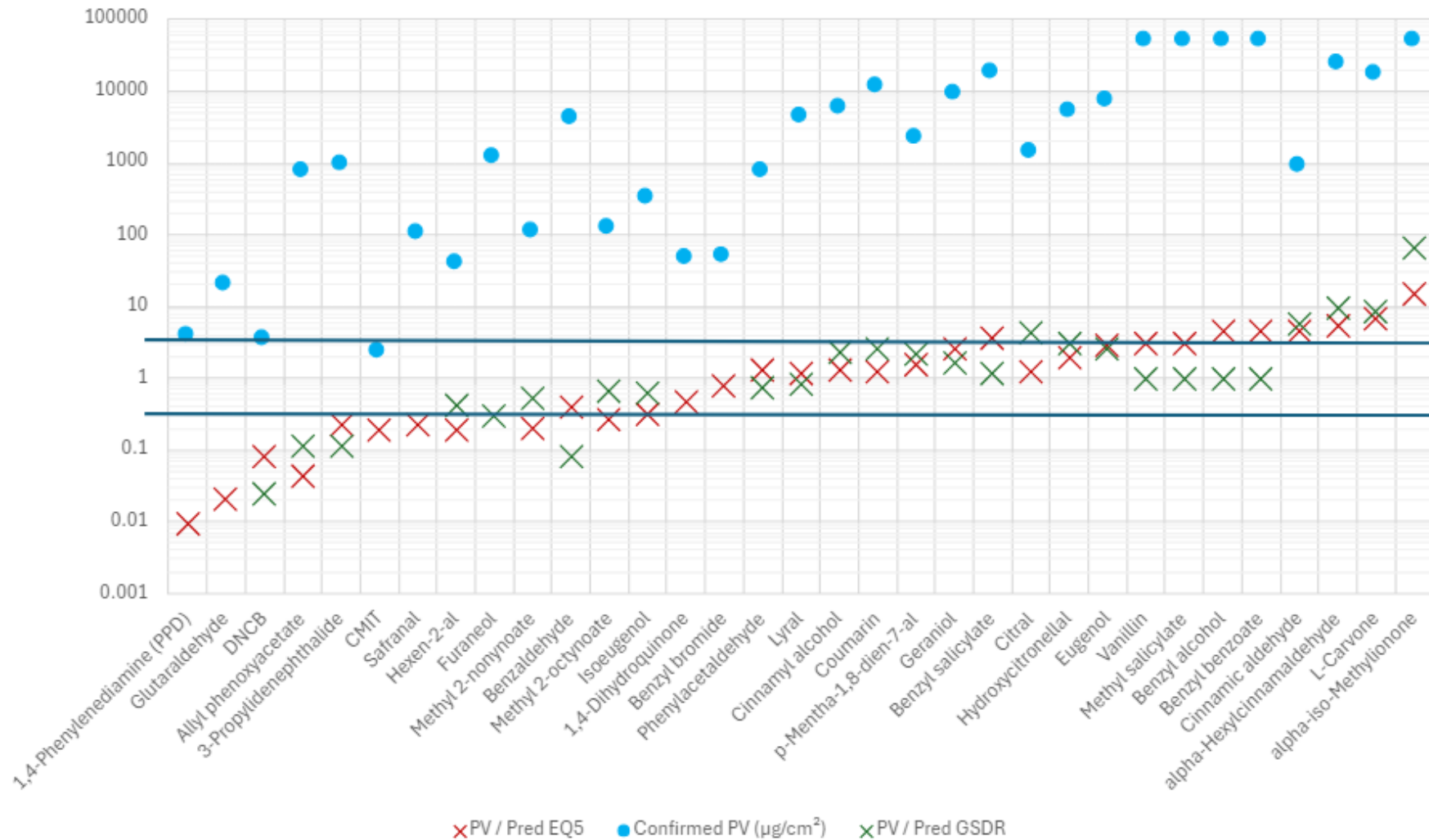
For analysis the ratio of the PV and either EQ5 or GSDR predictions is calculated

RCPL33 – PVs vs. Predictions

Ratio of PV and mean of EQ5 and GSDR – re-ordering



PV / Pred Mean Analysis



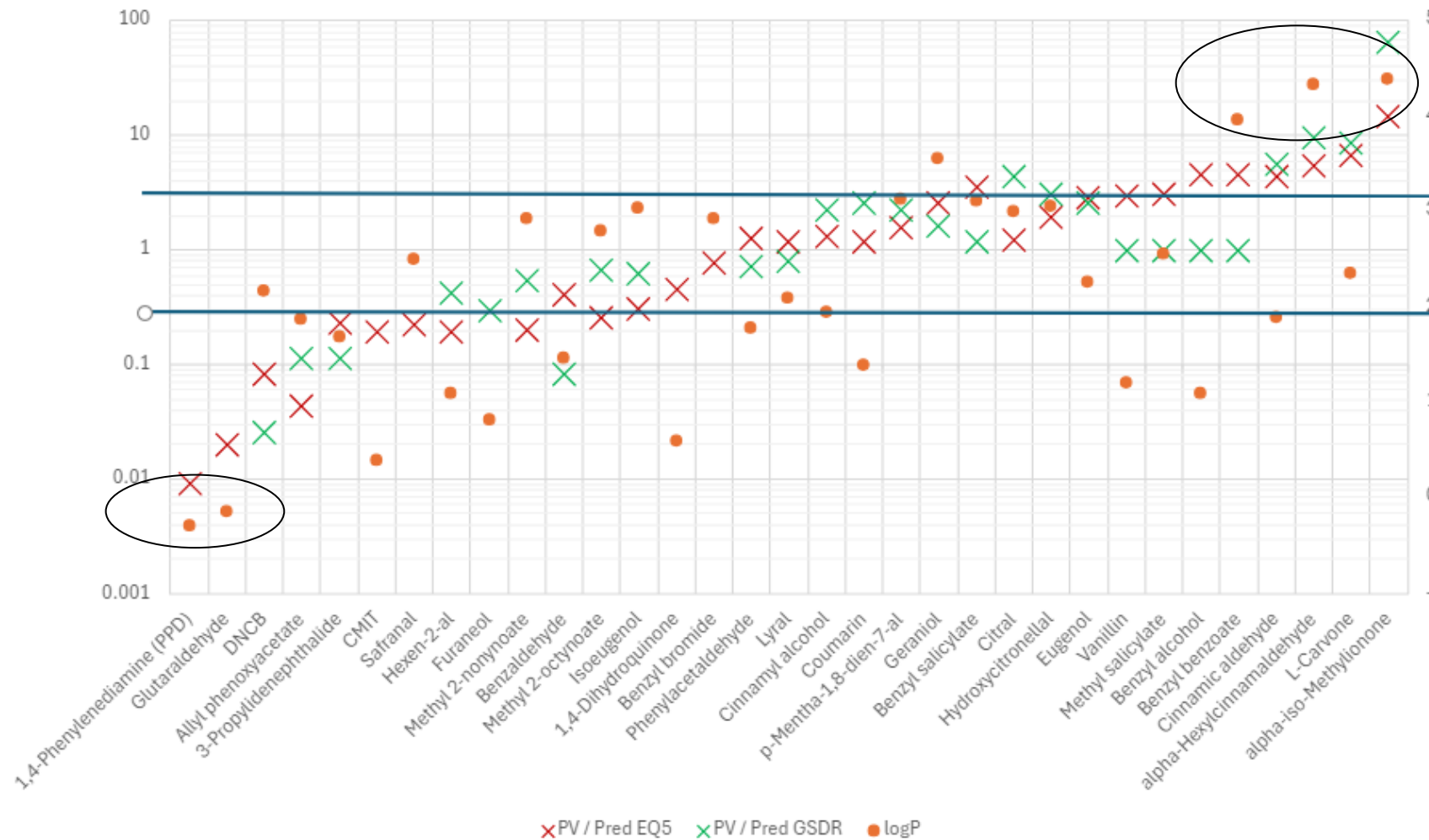
Re-ordering chemicals according to PV / EQ5 and PV / GSDR ratios

RCPL33 – PVs vs. Predictions

PV / Prediction ratio vs. LogP



PV / Pred Mean Analysis



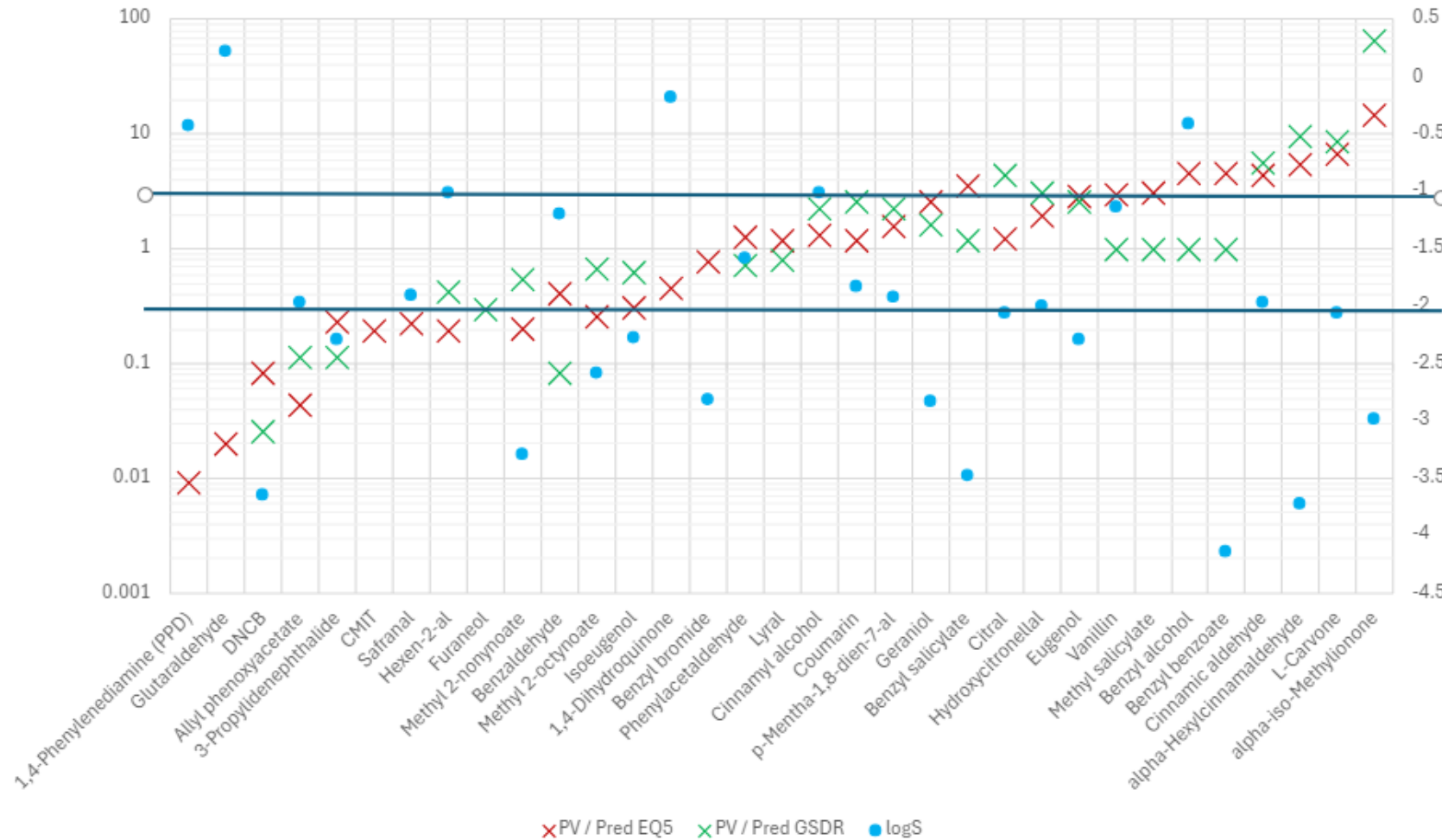
- Chemicals with very low logP values tend to show underprediction of their PV values
- Chemicals with very high logP values tend to show overprediction of their PV values
- Needs more chemicals to verify or falsify this observation

RCPL33 – PVs vs. Predictions

PV / Prediction ratio vs. LogS (water solubility)



PV / Pred Mean Analysis



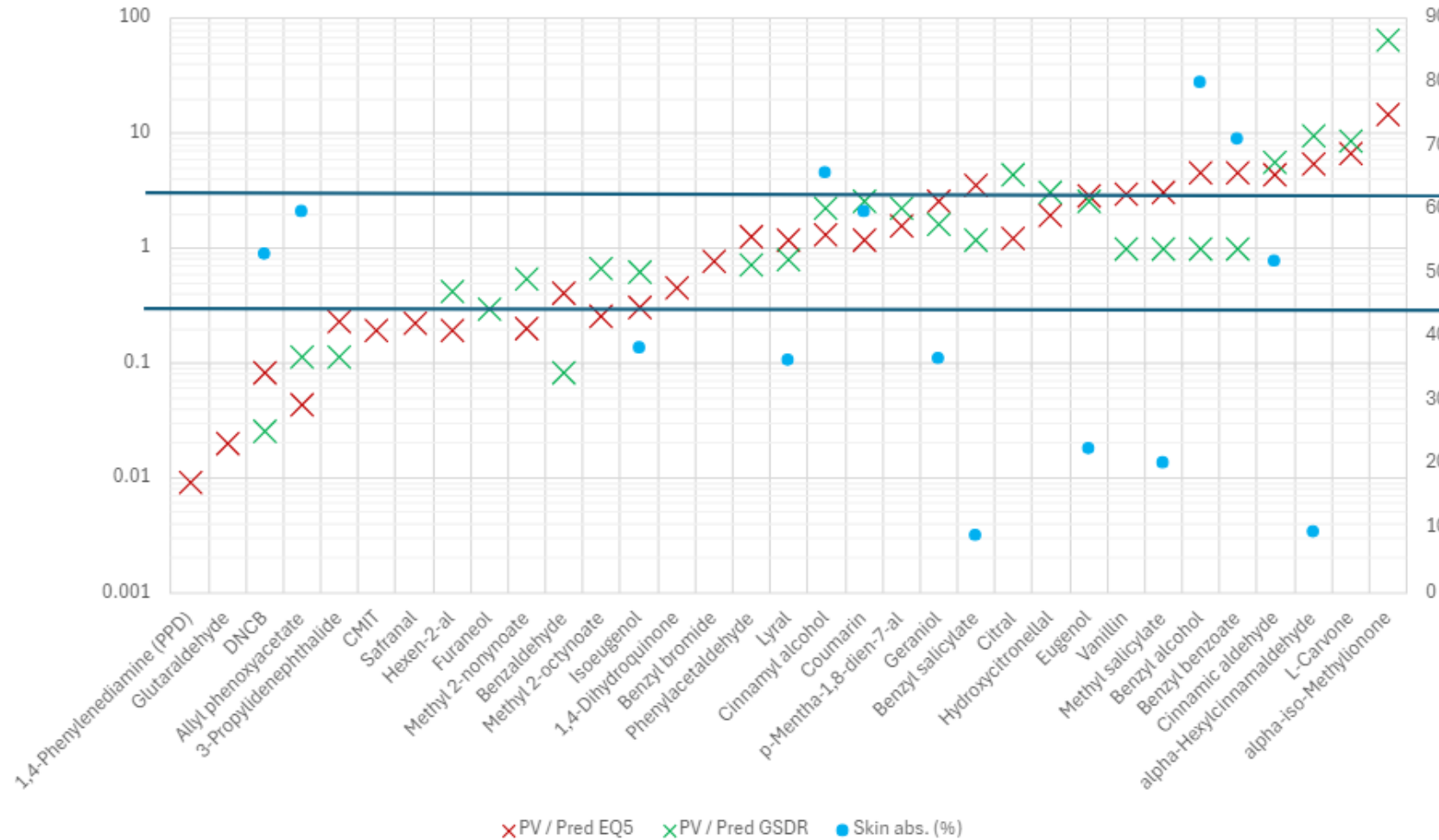
Water solubility seems to lack correlation with PV values

RCPL33 – PVs vs. Predictions

PV / Prediction ratio vs. measured skin absorption (%)



PV / Pred Mean Analysis



- Low skin absorption values tend to fall into area where NAMs tend to overpredict PV
- Needs more chemicals to substantiate this observation



symrise

*always
inspiring more*

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