where is the number of reactions metabolic genes: this is the theoretical maximum gene-expression penalty in some sense, although achieving this penalty would require either

for all ; for all ; and

or

for all ; for all ; and

in addition to the optimizer overriding the gene expression data for every single reaction metabolic gene.

Achieving this maximum penalty would require completely polarized aijs and a specific value of , among other things. Alternately, we could use a maximum penalty adjusted for – that is, the maximum penalty achievable given the value of , which would be

But this isn’t perfect either. For instance, if (so we’re only penalizing turning on low-expression reactions metabolic genes), theoretically , but this requires all the aijs to be 0 (so that ; see section “Gene Expression and Knockouts”) – not only perfectly confident, but also all predicted “off”. It might be better to also adjust for both and how many aijs are on each side of 0.5. That way, if (for instance) most of the aijs were above 0.5 and , would correctly reflect the fact that only a few genes are really capable of generating penalties (but then it would upweight those genes??? Is that what we want? For that matter, do we really want the scale changing as changes? Shouldn’t we leave the penalties alone regardless of aijs and , and just use the original definition? See reasoning for denying the third alternate definition, at the bottom)

[second alternate definition here]

This might be better; it would mirror in some sense the definition of as the maximum value of given these media and gene-knockout conditions.

Taking this further – which I do **not** recommend – would be to use a maximum penalty adjusted for both and the specific aijs; that is, the maximum penalty achievable given the values of and the aijs, which would be

The reason I do not recommend this is that then the normalized penalty, , would always have the same potential for impact on the objective function regardless of the level of confidence we have in the aijs. That is, the less overall confidence we have in the aijs, the more it artificially amplifies the penalties to compensate so that there is the same total potential for impact on the objective function. Instead, we *do* want the total potential for impact on the objective function to change based on the overall confidence level represented by the aijs, so we should *not* use this definition. However, we could use the maximum penalty adjusted for adjusted for (the first alternate definition proposed in this footnote).