Here is some data from the optimization Rose ran when she was working on adding M3 to the protocol (excel file attached).  The old protocol used 25 ng/well of OR, and she found better results when she used 4 ng/well instead.

My main issue with her optimization was that she tried to optimize for signal, rather than signal:noise.  I think one well per experiment is fine at first, but we should run technical replicates to make sure we aren't just amplifying the noise the same amount.

I have some older work where I use odd units that I can't work out without looking at the protocol.

I wrote:

HuOR275 concentration varied from 0.2-0.6 µg, while RTP1A1, Ric-8b, and Golf concentrations all varied from 0-0.2 µg.  Each transfection combination was then tested at three concentrations (no odor, 3 uM, and 30 uM) with two replicates.  Using JMP 6.0 I constructed a response surface to determine the optimum concentrations of the receptor proteins at which luciferase activity best distinguished between the three odorant concentrations.  Two-dimensional projections of the response surface are presented in figure 4 (tif attached).  This analysis determined that the optimal concentrations were 0.6 µg of HuOR275, 0.09 µg of RTP1A1, 0.07 µg of Ric8b, and 0.04 of µg Golf.

Hope you can make sense of this...

Best,

Joel