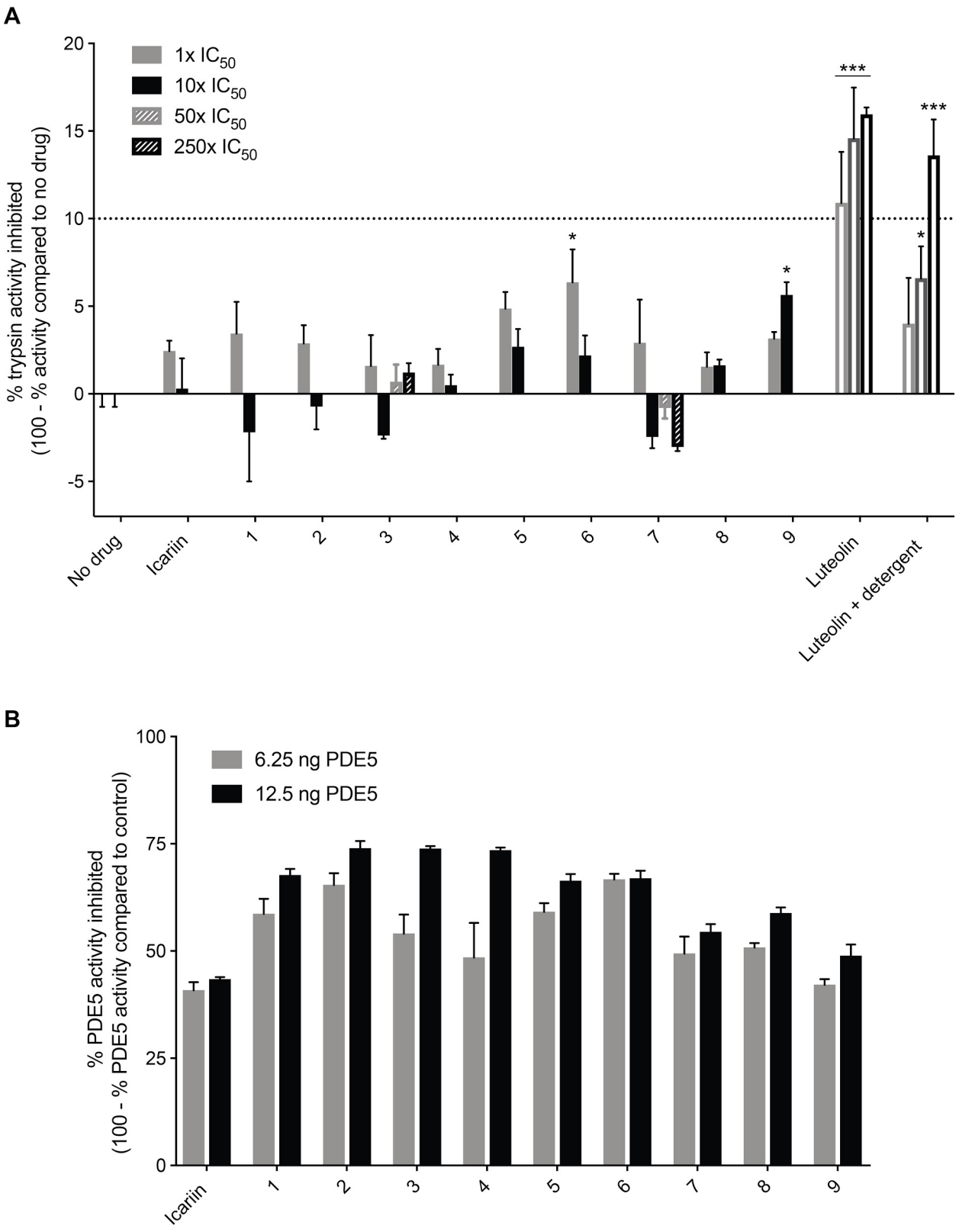

S10 Fig. Icariin and compounds 1-9 do not show characteristics of pan-assay interference compounds (PAINS). (A) Compounds were assayed for trypsin inhibition at 1x

and 10x the PDE5 IC_{50} concentration. Compounds **3** and **7** were also assayed at 50x and 250x the IC_{50} concentration. Luteolin was assayed at 50, 100, and 250 μM (light grey, dark grey, and black outlined bars respectively). The addition of detergent is expected to reduce the non-specific inhibitory effects of PAINS compounds. The decrease in trypsin inhibition by luteolin in the presence of detergent further confirms luteolin as a PAINS compound. Values were measured in duplicate. Data is expressed as mean ± SEM. Average trypsin reaction velocity in no drug control reactions: 236 ± 2 nmol min^{-1} mg^{-1}. *p<0.05, ***p<0.001 *vs* no drug controls (one-way ANOVA with Dunnet's multiple comparisons test). (B) Icariin analogs at 1x the PDE5 IC_{50} concentration were assayed against a 2x increase in PDE5 concentration. Data was measured in triplicate and normalized to the untreated control median. Data is expressed as mean ± SEM. Average PDE5 reaction velocity in no drug control reactions: 1.50E-7 ± 4E-9 μmol/min with 6.25 ng PDE5; 3.4E-7 ± 1E-8 μmol/min with 12.5 ng PDE5. The difference between the mean of 6.25 ng PDE5 and the mean of 12.5 ng PDE5 was not statistically significant for any treatment (*t* test corrected for multiple comparisons with the Holm-Sidak method). .