

Supplemental information

6-Thioguanine blocks SARS-CoV-2

replication by inhibition of PLpro

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Supplemental Figure 1 related to Figure 1

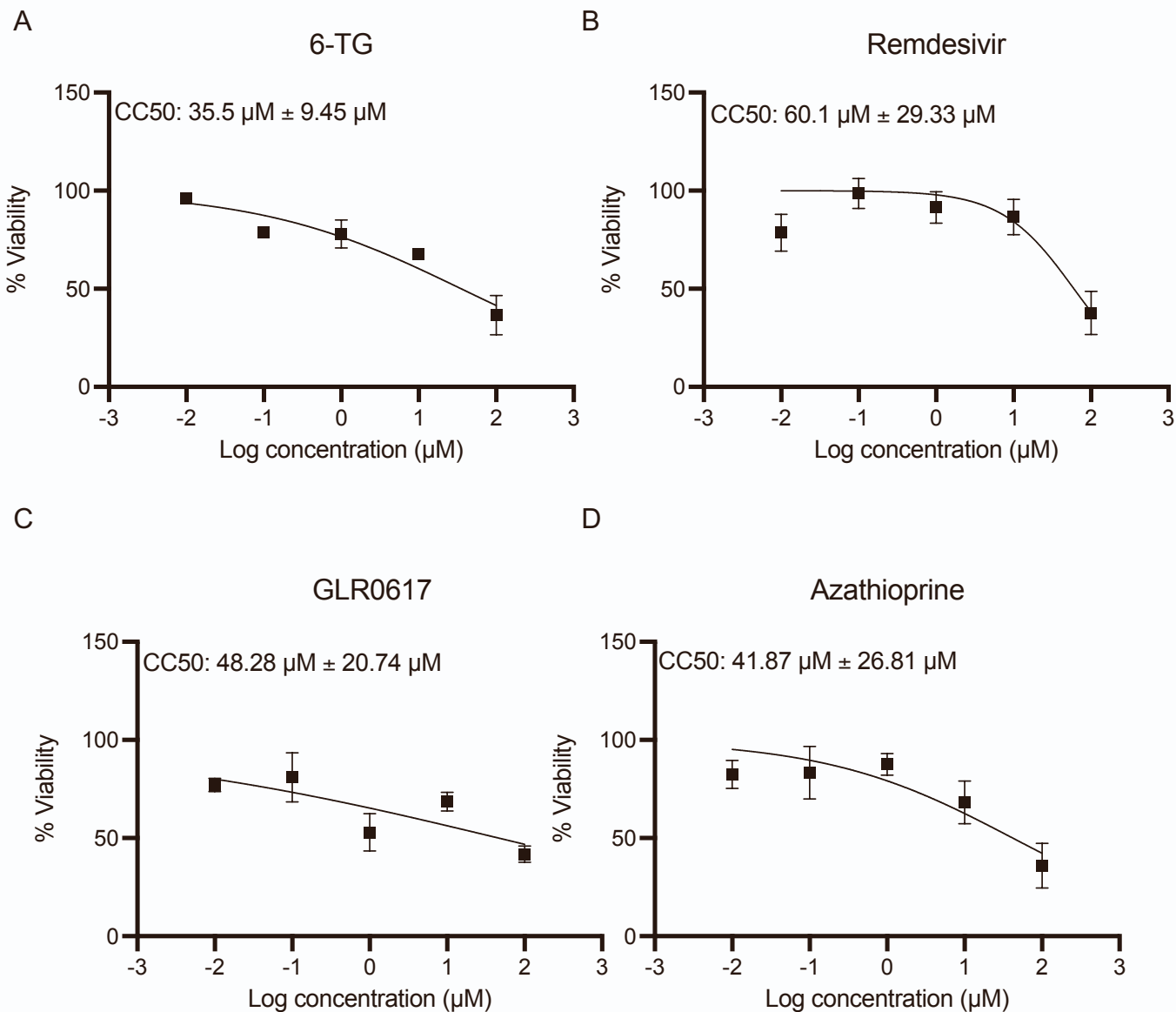


Figure S1, related to Figure 1. Compound cytotoxicity assays. The compounds were tested for cytotoxicity using the CellTiter-Glo 2.0 kit (Promega). Vero-E6 cells were treated in triplicate for 48 hours with increasing concentrations (.01µM, .1µM, 1µM, 10µM, and 100µM) of the compounds and CC50 was determined using Graphpad Prism 9 software. A. 6-TG: CC50 35.35µM B. Remdesivir: CC50 60.10µM C. GLR0617: CC50 48.28µM and D. Azathioprine: CC50 41.87µM. 95% confidence limits are indicated for each.

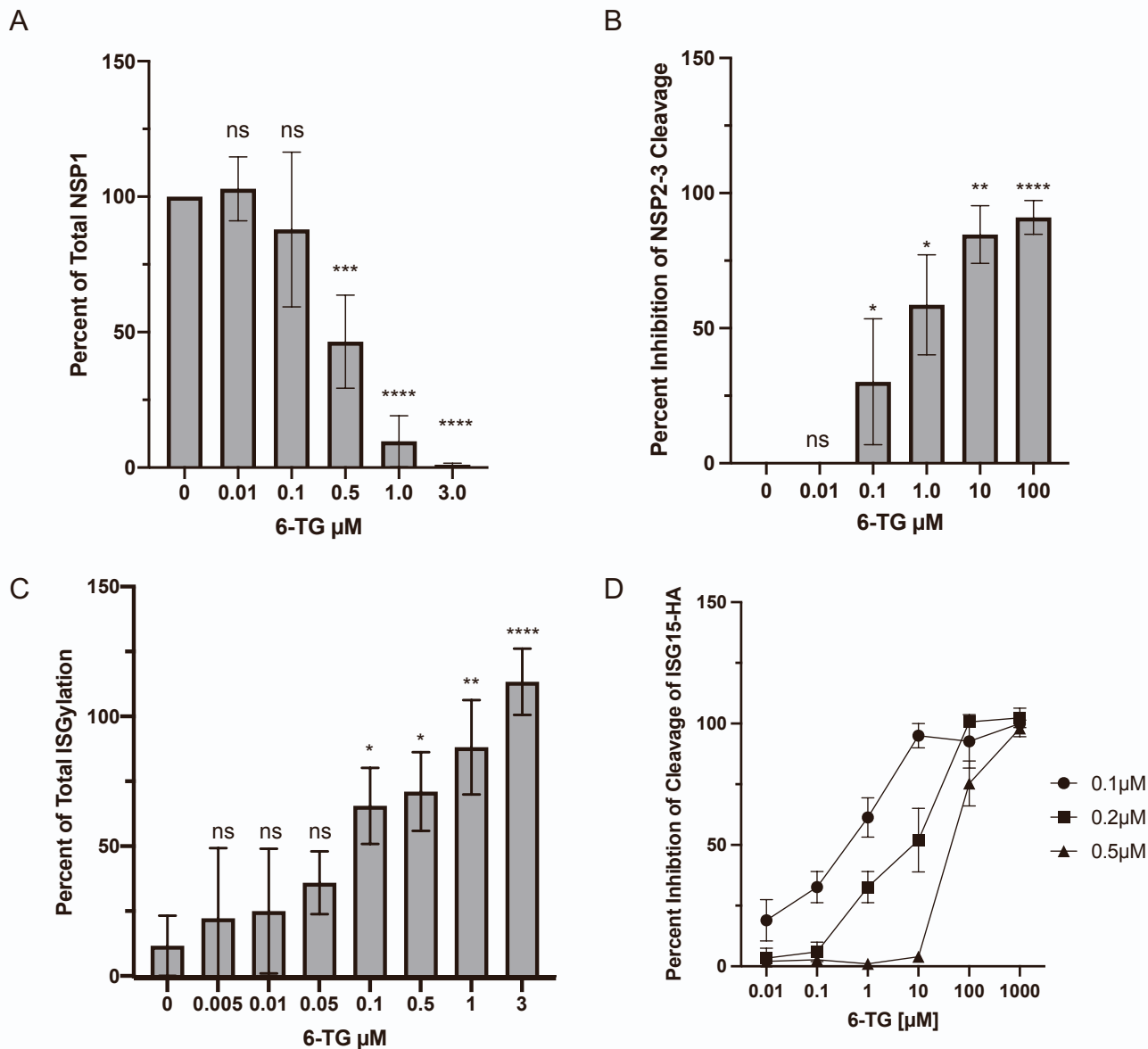


Figure S2, related to Figures 2 and 3. Quantification of PLpro activities. A. Densitometry analysis of TAP-nsp1 from Figure 2A. Percent TAP-nsp1 was determined by comparison to the 42 kDa band in the untreated samples to those present in samples treated with 6-TG. B. Densitometry analysis of TAP-nsp1 from Figure 2B. Percent Inhibition of TAP-nsp2-3 cleavage was determined by comparison to the ~40 kDa TAP-nsp2 band in the untreated samples to those present in samples treated with 6-TG. C. Densitometry analysis of ISGylation from Fig. 3A. Percent of total ISGylation was determined by comparison to total ISGylation in the presence of PLpro^{CA}. D. Densitometry analysis of ISG15-HA cleavage from Fig. 3B. Percent of inhibition of cleavage of ISG15-HA was calculated by comparing the -PLpro sample with the samples treated with PLpro and 6-TG. Significance was assessed by ordinary one-way ANOVA comparison to treatment with no 6-TG. Asterisks indicate p-values: *=0.02, **=0.002, ***=0.004, ****<0.0001, and non-significant changes are indicated by ns.