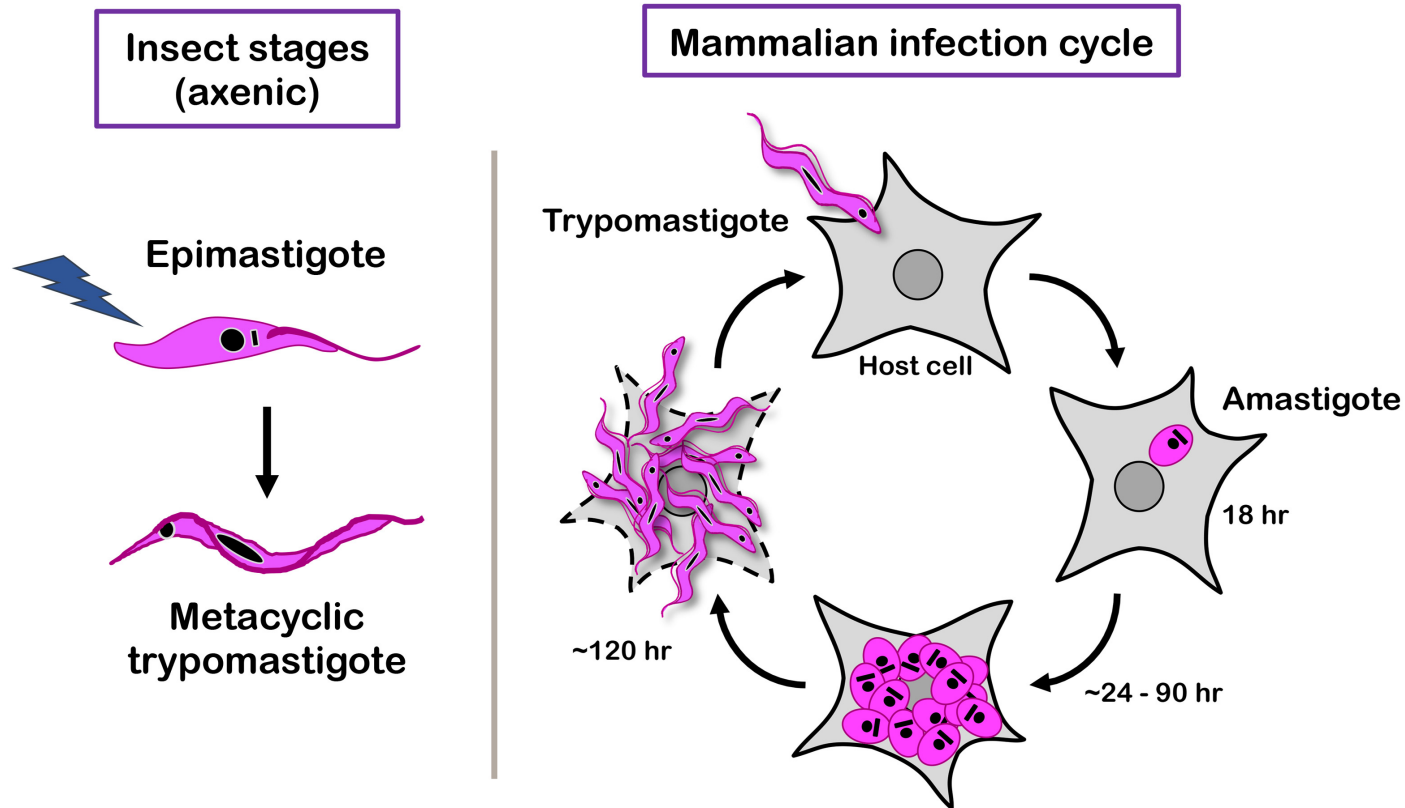
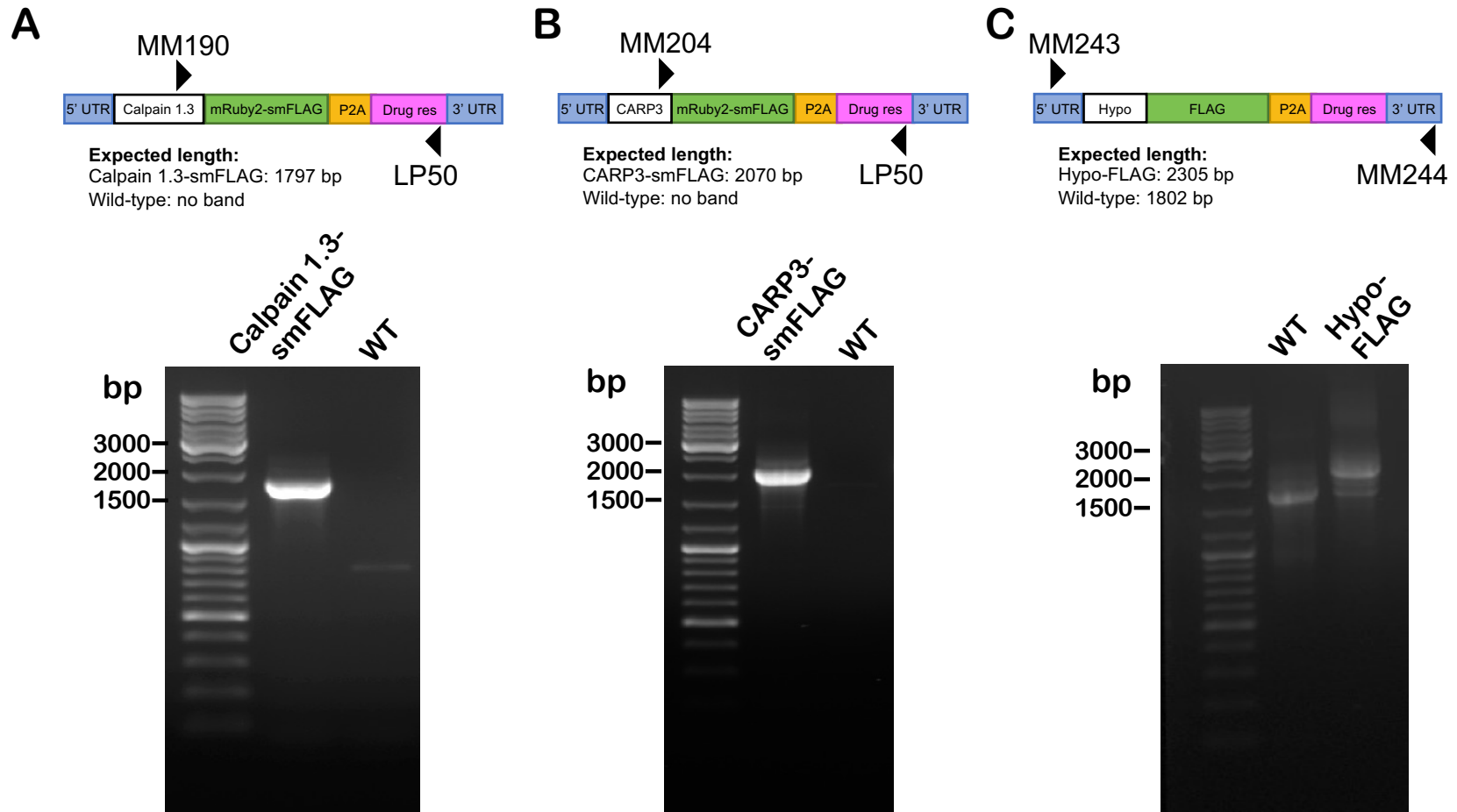


## Supplemental Figure 1: Won et al.



**Supplemental Figure 1. Schematic of the *T. cruzi* life cycle.** Insect stage '*epimastigote*' that is propagated axenically in liquid culture and gives rise to the infectious '*metacyclic trypomastigote*'. Trypomastigotes, whether derived from epimastigotes or as the end product of a single lytic cycle in a mammalian cell are motile, non-dividing forms of the parasite that actively invade a mammalian host cell. Inside a host cell, the '*trypomastigote*' transforms into the replicative intracellular '*amastigote*' stage by 18 hours post-infection (hpi). Amastigotes undergo several rounds of proliferation, dividing by binary fission (between ~24-90 hpi), before they stop dividing and differentiate into trypomastigotes, that eventually lyse the infected host cell and disseminate infection. Stable transfection and drug selection is performed in the stage (lightning bolt symbolizes electroporation). Once stable genomic changes are confirmed in epimastigotes, these parasites are used to establish the mammalian infection cycle starting with metacyclic trypomastigotes as outlined above.

## Supplemental Figure 2: Won et al.



**Supplemental Figure 2: PCR confirmation of endogenous tags for candidate proteins.** **A.** Schematic showing the region of amplification and DNA gel with corresponding bands for **A.** calpain 1.3-smFLAG (TcCLB.506563.200), **B.** CARP3-smFLAG (TcCLB.506681.40), and **C.** hypothetical protein-FLAG (TcCLB.510329.180). DNA ladder shown with sizes (bp) on left.