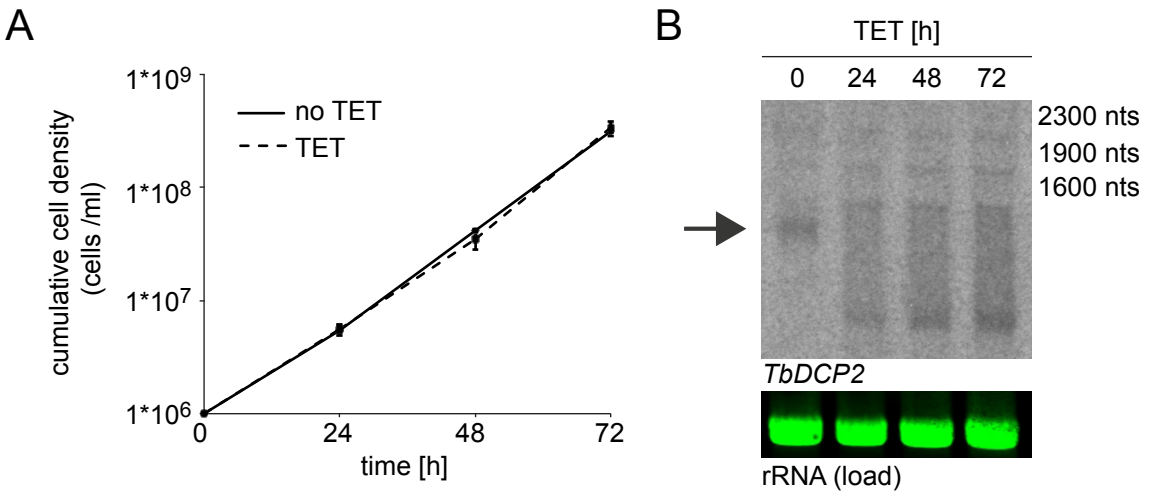

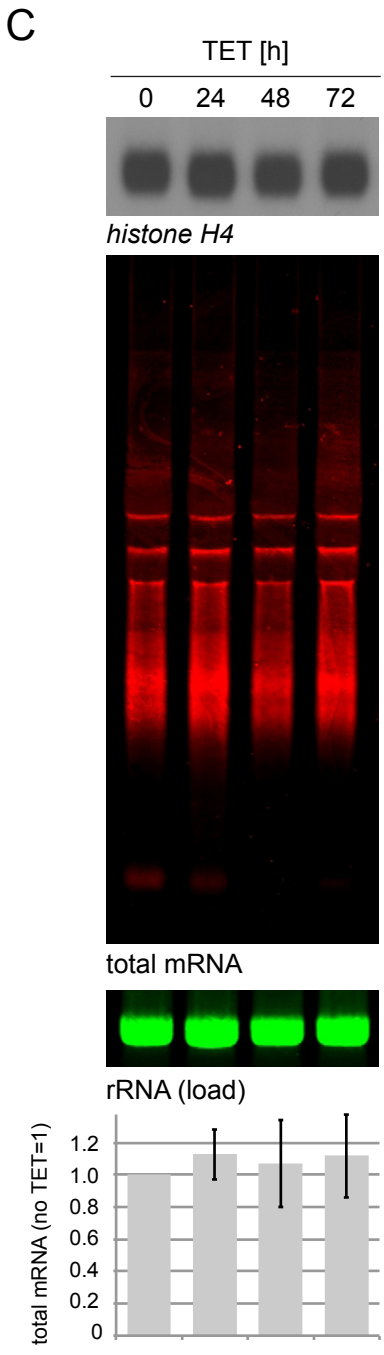

Figure S11: RNAi of TbDCP2 (Tb927.6.2670)
The C-terminal 698 nucleotides of the DCP2 open reading frame were cloned head to head into p3666 (Sunter et al., 2012). After transfection into PSPR2.1 cells (Sunter et al., 2012) the expression of a stem loop RNA is inducible by tetracycline (TET). Three independent RNAi cell lines were analysed.

A) Growth of three independent clonal RNAi cell lines was measured in the absence (no TET) or presence (TET) of tetracycline. No growth effect was detected. The error bars represent standard deviations between the three clonal cell lines.

B) Northern blot to show the RNAi induced reduction in *DCP2* mRNA. RNA samples were collected over a time-course of DCP2 RNAi and analysed by a northern blot probed for *DCP2* and for rRNA (loading control). The expected size of the *DCP2* mRNA (arrow) is 1068 nucleotides. The mRNA becomes undetectable after 24 hours of RNAi induction; instead mRNA bands of smaller size are visible, possibly products of the RNAi pathway. This is a representative northern for one RNAi clone, the northern blots of the other two RNAi clones look similar.

C) Northern blots with mRNA collected over an DCP2 RNAi time-course probed for *histone H4* (top), for total mRNA with an oligo antisense to the miniexon sequence (middle) and for rRNA as a loading control (bottom). One representative northern blot of one RNAi clone is shown; the northern blots of the other two RNAi clones look similar. The amount of total mRNAs was quantified; average values of the three RNAi cell lines are shown with standard deviations represented as error bars. We observed no change in the length of the poly(A) tail for histone H4 mRNA (compare with Figure 3 for controls), and no change in the amount of total mRNA.

Sunter J, Wickstead B, Gull K & Carrington M (2012) A new generation of T7 RNA polymerase-independent inducible expression plasmids for Trypanosoma brucei. PLoS ONE 7: e35167