ABSTRACT

Chromosomal translocations result from joining of DNA double-strand breaks (DSBs) and frequently cause cancer. Yet, the steps linking DSB formation to DSB ligation remain undeciphered. We report that DNA replication timing (RT) directly regulates lymphomagenic *Myc* translocations during antibody maturation in B cells downstream of DSBs and independently of DSB frequency. Depletion of minichromosome-maintenance (MCM) complexes alters replication origin activity, decreases translocations, and deregulates global RT. Ablating a single origin at *Myc* causes an early-to-late RT switch, loss of translocations, and reduced proximity with *Igh*, its major translocation partner. These phenotypes were reversed by restoring early RT. Disruption of early RT also reduced tumorigenic translocations in human leukemic cells. Thus, RT constitutes a general mechanism in translocation biogenesis linking DSB formation to DSB ligation.