

Abstract

Faithful duplication of DNA is crucial for maintenance of genomic stability and viability of cells. The initial event of DNA replication is characterized by the binding of the pre-replication complex (pre-RC) onto origins of replication. Pre-RCs are licensed for replication by the loading of the licensing factor CDT-1, which promotes the subsequent recruitment of several other replication factors, including CDC-45 and GINS, to form an active replisome and thereby enable DNA synthesis. To hinder re-assembly of pre-RCs and thus re-initiation of DNA replication within the same cell-cycle central replication factors are targeted for degradation. Recent findings identified a pivotal involvement of the AAA ATPase CDC-48 (also known as p97 in vertebrates) in the regulation of DNA replication. CDC-48 is known to mediate mobilization and targeting of ubiquitylated proteins to the 26S proteasome. Studies in *C. elegans* revealed an essential function of a complex, consisting of CDC-48 and its substrate-recruiting cofactors UFD-1 and NPL-4, in the targeting and mobilization of the DNA replication factors CDT-1 and CDC-45/GINS from chromatin. In *C. elegans* embryos, deficient for a functional CDC-48^{UFD-1/NPL-4} complex, CDT-1 and CDC-45/GINS are misregulated and associated persistently with chromatin, which consequently results in severe DNA replication stress and eventually in embryonic lethality.

However, the detailed function of CDC-48, and particularly its cofactors, during DNA replication processes is still opaque. This work aimed to elucidate the functions of the CDC-48 cofactor UFD-1 during DNA replication events. Additionally, this work describes the previously unidentified role of the CDC-48 cofactor UBXN-3 in the regulation of the DNA replication licensing factor CDT-1. It is shown that, upon the depletion of a functional CDC-48^{UBXN-3} complex, *C. elegans* embryos stabilize CDT-1 on mitotic chromatin. In combination with the finding of a direct interaction of UBXN-3 with CDT-1, this suggests a UBXN-3-dependent recruitment of CDC-48 to chromatin, which is required to maintain genomic stability. Furthermore, worms deficient for *ubxn-3* were identified to have a reduced lifespan upon hydroxyurea treatment, which potentially links DNA replication stress with impaired longevity.