Abstract

Giardia intestinalis is a protozoan causing diarrhea worldwide. Beside its medical importance, it is evolutionary distant protist with two nuclei within a cell adapted for parasitic life in the environment poor of oxygen. Its genome is small and compact in term of gene content and size. It is therefore an attractive model organism for studies of minimal requirements for cellular processes. Present work brings new partial information on different levels of chromosome integrity maintenance of this parasite.

Our study presents characteristics of chromosome termini and their protection. We localized telomeres during all stages of the trophozoite cell cycle and determined the length of *Giardia* telomeres ranging from 0.5 to 2.5 kb, we proved an existence of an active telomerase enzyme synthesizing telomeric repeats in in this parasite, despite the fact that giardial telomerase is structurally divergent. Present data support the view that the chromosomal termini in *Giardia* are maintained in a conservative manner that is common to other eukaryotes.

We described effects of commonly used drug for treatment of anaerobic infections, metronidazole, on DNA and cell cycle progression in susceptible and resistant cell lines. Incubation of cells with this drug causes phosphorylation of histone H2A in cell nuclei and fragmentation of DNA. Sublethal concentration affects the replication phase of the cell cycle and lethal drug concentration lead to rapid loose of adherence ability without any effect on cell cycle progression. Our results support the view that the early reaction of cells to lethal concentration of metronidazole is not primarily initiated by the reaction to DNA damage but rather by the immediate interaction of the drug with biomolecules where active form of metronidazole is generated. In resistant lines incubated in the presence of the drug, about 40% of cells remain permanently positive for H2A in nuclei without any effects on the cell cycle progression. This suggests that DNA damage caused by this drug treatment persists in these cells and may contribute to accelerated mutagenesis and consequently to the development of natural resistance.

Chromosomes in *Giardia* condense, we described the overall morphology, condensation stages, and mitotic segregation of these chromosomes, which is similar to other model eukaryotes. Differently, the anaphase poleward segregation of sister chromatids is atypical and

tends to generate lagging chromatids between daughter nuclei which could explain existence of aneuploidy in this parasite. On molecular level, *Giardia* lacks several genes involved in the cohesion and condensation pathways, in present study we identified two putative members of the kleisin family thought to be responsible for condensin ring establishment.

Lastly, we examined effects of synchronization agent aphidicolin on the nuclear cycle and cell cycle progression characteristics, as well as their reversibility. Treatment with aphidicolin leads to G1/S phase arrest and to phosphorylation of H2A histone. Thus, if aphidicolin is used for synchronization of *Giardia* trophozoites, this fact must be accounted for, and treatment with aphidicolin must be minimal.