

Nuclear genes of euglenids contain two types of introns: conventional spliceosomal introns with GT/C-AG borders and nonconventional introns. Nonconventional introns have non-canonical borders and form a stable RNA secondary structure bringing together both ends. Conventional introns are present at conserved positions, while nonconventional ones at positions unique to individual species or clades grouping closely related taxa. Some introns (intermediate introns) have features of both types and it is expected that they could be a transitional form between the conventional and nonconventional introns. Knowledge on the acquisition of new nonconventional introns is limited to data for individual genes. The best source of larger data sets seems to be sequencing of nuclear genomes of different representatives of euglenids. Therefore the main objective of the project is sequencing of nuclear genomes of *Euglena hyemalis* and *Euglena longa*. The analysis of introns' sequences and introns' distribution in homologous genes will allow for: (1) estimation of the scope of intron gains and losses, (2) estimation of the number of new nonconventional introns acquired after the split of evolutionary lineages *E. longa* - *E. hyemalis*/*E. gracilis* and *E. hyemalis* - *E. gracilis*, (3) estimation of most conserved elements within nonconventional introns based on the large amount of data, (4) comparison of the sequence/structure of old nonconventional introns (present in all species) and newly acquired ones (unique for *E. hyemalis* or *E. gracilis*), which could help to identify elements characteristic for the source sequence of nonconventional introns and also for mechanism of introns acquisition, (5) identification of intermediate introns excised by two different mechanisms, (6) identification of introns for which the change of the type/mechanism of excision took place.

Two next generation sequencing technologies will be applied: Illumina and PacBio methods. Predicted gene models will be compared for two genomes obtained in the project and also for genome of *E. gracilis*. Homologues will be identified, then comparative analyses of introns distribution, as well as analyses of the sequence and structure of nonconventional introns will be carried out.

The analysis of nuclear genomes of euglenids will allow for global comparison of introns distribution in different species and for searching of intermediate introns. Genomic data will also enable more detailed analysis of conserved features of nonconventional introns, especially those that have been inserted relatively recently. It would help to identify their origin and the mechanism of insertions. The obtained data on evolution of nonconventional introns in nuclear genes of euglenids will contribute to expanding our knowledge about the role of intervening sequences in eukaryotic genes, which is still not completely understood.