

**The mitochondrial genome of moss *Brachythecium rivulare* B.S.G.
(Hypnales, Brachytheciaceae)**

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The liverworts and mosses (bryophytes *sensu lato*) are the basal groups of higher plants diverged more than 450 million years ago (myr). The primary terrestrial biotopes formed by bryophytes were an important spots for subsequent invasion on land of other plants. To date there is no comprehensive scenario of this crucial step of plants evolution. Now, one possible way to clarify some evolution obscurities is a comparative genomics approach. However, until now bryophyte genomics is on some initial stage of its progress especially in comparison with other groups of plants. To date, the nuclear genome sequence as two scaffolds available for a single moss species - *Physcomitrella patens*, and for two species plastid genomes are known. Mitochondrial genomes also from only two moss species from different subclasses Bryidae and Funariidae are deposited in NCBI GenBank: *Anomodon rugelii* (Hypnales, Anomodontaceae) and *P. patens* (Funariales, Funariaceae).

Taking into account the great diversity of bryophytes (in number of species, they are not inferior angiosperms) from one hand and scanty bryophyte mitochondrial genome data on the other hand, we sequenced and annotated mitochondrial genome of *Brachythecium rivulare* (as a part of whole genome sequencing project for this species). About 1 µg of total DNA was

extracted from *B. rivulare* plant sample. Two pair-end libraries with 164 nt and 259 nt were performed. The sequencing procedure was accomplished on NGS platform Illumina HiSeq 2000 in two flow cell lines. Both library preparation and sequencing were performed following standard Illumina protocols.

The raw sequencing data was comprised approximately 187 and 175 million of 101 nt paired reads, respectively. After trimming of low quality read positions and sequencing adapters removal-10 million read pairs was extracted from each library and assembled using Velvet (with *k*-mer length 91). The assembly consisted of 617 contigs with total length of 573364 nt; the longest one - 104,474 nt in length - was the complete mitochondrial genome with partially overlapping ends and 61x coverage. The correctness of closing of linear sequence in circular was verified by mapping initial reads subset to this sequence without overlapping ends by Bowtie 2.

The size of circular mtDNA molecule is 104,460 nt for *B. rivulare*, 105,340 nt for *P. patens*, and 104,239 nt for *A. rugelii*. Mitochondrial genome of *B. rivulare* consists of the same genes situated in the same order as in two other mosses. The difference in size attributed mainly to changes in the length of non-coding regions. It seems that overall structure of mitochondrial genome of mosses is significantly more conservative compared with angiosperms where its gene content and size may vary tremendously even for species of a single family. Although the time of angiosperm divergence is significantly less the diversification time of extant groups of mosses. The diversification time of moss lineages calculated as ca. 405 myr, the split of Bryidae and Funariidae as ca. 224 myr, and divergence of *B. rivulare* and *A. rugelii* occurred roughly 114 myr ago; the split between Magnoliales and monocots was set at 123 myr [1].

The mean similarity of ungapped mtDNA sequence from *B. rivulare* and *A. rugelii* is 96.3%, and from *P. patens* and two other species is 84.7%. These values correspond to differences in the age of these species.

1. Newton A.E., Wikstrom N., Bell N., Forrest L.L., Ignatov M.S. (2007) Dating the diversification of the pleurocarpous Mosses. In: Pleurocarpous Mosses. Systematics and Evolution, Newton A.E. and Tangey R.S. (Eds.) 337-366 (CRC Press).