
Comparison of Mitochondrial Genomes of Bryophytes

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Abstract: Bryophytes (liverworts, mosses and hornworts) hold the basal most position among extant land plants. The present study deals with comparative analysis of available mitochondrial genomes of bryophytes using bioinformatics tools. These mitochondrial genomes belong to 2 liverworts, 1 moss and 2 hornworts. The analysis is based on multiple genome analysis through whole proteome comparison to detect conservation between mitochondrial genomes of bryophytes.

Introduction

Bryophytes hold a basal position in the phylogeny of land plants. The first land plants were considered to have a bryophyte type of developmental organization (Knoop 2010). The advent of automated DNA sequencing technologies helps to generate molecular data for various plant species including bryophytes. Molecular data analysis assists to build hypotheses of early land plant origins. Moreover it supports to establish phylogenetic relationship of plant lineages. The three distinct clades of bryophytes (liverworts, mosses and hornworts) were supported as paraphyletic in origin (Dombrowska and Qiu 2004; Kelch *et al*, 2004; Groth-Malonek *et al*, 2005; Wolf *et al*, 2005; Qiu *et al*, 2006; Gao *et al*, 2010). Contrary to this several studies showed monophyly of bryophytes (Garbary *et al*, 1993; Nishiyama *et al*, 2004; Goremykin and Hellwig 2005; Shanker *et al*, 2011). Most of these studies used chloroplast and/or mitochondrial genome sequences to infer phylogenetic relationship of extant bryophytes.

In comparison to genome sequencing of a large number of chloroplast genomes, few mitochondrial genomes of plants have been sequenced which belong to various algae, bryophytes, pteridophytes, gymnosperms and angiosperms (NCBI Eukaryotae Organelles List). The availability of complete mitochondrial genome sequences of plants makes it feasible to compare these genomes and detect conservation among them. Moreover it would help to establish phylogenetic relationship and possible course of plant evolution.

Earlier, sequence similarity search tools with specific search criterion were used on chloroplast genomes of bryophytes (Shanker and Sharma 2012). These similarity search tools identify orthologous sequences among organisms. Orthologous sequences provide insight on conservedness among organisms utilizing the treasure of evolutionary information present in complete genome sequences. In the present study a data set of mitochondrial proteomes of bryophytes was used to detect conservation between them.

Materials and Methods

Mitochondrial Genome Sequences

Mitochondrial genomes of bryophytes lineages available in National Center for Biotechnology Information (NCBI) organelles database were used in this study. A total of 5 mitochondrial genomes were found in the database. These mitochondrial genomes belong to 2 liverworts, *Marchantia polymorpha* (Oda *et al*, 1992) and *Pleurozia purpurea* (Wang *et al*, 2009), 1 moss, *Physcomitrella patens* (Terasawa *et al*, 2007) and 2 hornworts, *Phaeoceros laevis* (Xue *et al*, 2010) and *Megaceros aenigmaticus* (Li *et al*, 2009). The protein sequences of these mitochondrial genomes were retrieved from NCBI. The accession number of mitochondrial genomes and the number of protein sequences downloaded is shown in table 1.

Comparison of Mitochondrial Sequences

All the surveyed proteomes were compared using protein-protein BLAST (Basic Local Alignment Search Tool; Altschul *et al*, 1997) to identify orthologous sequences (Genes/proteins separated by speciation) between them. During comparison the proteome of an organism once act as query proteome whereas at the same time other proteomes remain in the database. BLAST results were parsed using stringent measure of both sequence coverage ($\geq 50\%$) and percent sequence similarity ($\geq 30\%$). Parsed hits were used to identify putative reciprocal orthologous sequences.

Table 1. Information of Mitochondrial Genomes used in study.

S. No.	Organism Name	Accession Number	No. of Proteins
Liverworts			
1.	<i>Marchantia polymorpha</i>	NC_001660	76
2.	<i>Pleurozia purpurea</i>	NC_013444	69
Moss			
3.	<i>Physcomitrella patens</i>	NC_007945	42
Hornworts			
4.	<i>Phaeoceros laevis</i>	NC_013765	38
5.	<i>Megaceros aenigmaticus</i>	NC_012651	48

Results and Discussion

The results of identified orthologs among mitochondrial genomes of bryophytes are presented in table 2. Considering specified criteria of sequence positivity the proteome comparison between

organisms detect putative orthologs. The comparison of total N organisms leads to an $N \times N$ matrix. The matrix cells contain total number of putative orthologs identified between organism A and organism B. The diagonal of this matrix does not contain any values since these cells correspond to comparison with itself. The mitochondrial genome comparison of liverworts shows higher number of orthologs identified with mosses. This finding is in strong agreement with several earlier studies which showed liverworts and mosses as sister clades (Qiu *et al*, 2006, 2007; Qiu 2008; Chang and Graham 2011). Moreover phylogenetic analysis of bryophytes also showed liverworts and mosses forming a joint clade (Nickrent *et al*, 2000; Nishiyama *et al*, 2004; Shanker *et al*, 2011). Previously the position of hornworts was resolved as sister to vascular plants (Qiu *et al*, 2006; Pena *et al*, 2008). Moreover a recent analysis on chloroplast genomes of bryophytes and pteridophytes using newly defined bioinformatics parameters (Shanker *et al*, 2009) identified hornworts as transition link between bryophytes and pteridophytes (Shanker and Sharma 2012). Both representatives of hornworts used in this study showed equal number of orthologs with liverworts and mosses. However the number of orthologs found with liverworts and mosses are very less (table 2). This clearly indicates that due to evolutionary distance between these organisms the sequences become much diverged so that less sequence similarity was found.

Table 2. Putative reciprocal orthologs identified among mitochondrial genomes of bryophytes.

Organism	<i>Marchantia</i>	<i>Pleurozia</i>	<i>Physcomitrella</i>	<i>Phaeoceros</i>	<i>Megaceros</i>
<i>Marchantia</i>	-	45	41	20	21
<i>Pleurozia</i>	45	-	40	20	21
<i>Physcomitrella</i>	41	40	-	20	21
<i>Phaeoceros</i>	20	20	20	-	21
<i>Megaceros</i>	21	21	21	21	-

The sequence comparison of mitochondrial protein sequences of bryophytes shows conservation between these genomes. The orthologous sequences detected in this analysis is in strong agreement with the fact that more distant organisms show less sequence similarity in comparison to closely related organisms.

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