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Update section

Sequence

An equivalent to bacterial *ompR* genes is encoded on the plastid genome of red algae

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The understanding of the complex processes underlying the evolution and maintenance of a eucaryotic cell with organelles depends on the availability of data from organisms of different systematic positions. The comparison of unquestionably less related taxa might enable us to reconstruct the evolutionary history of organisms and to find common rules of cooperation between subcellular compartments, such as mitochondria or plastids and the nucleus.

The plastid genomes of chlorophytic, chromophytic and red algae share some common features: the genome size is insufficient to encode all proteins the organelle needs, so it has to import nucleus-encoded proteins from the cytoplasm. All plastid genomes investigated so far encode the genes for the ribosomal RNAs, a set of tRNAs and genes for essential proteins of the photosynthetic apparatus such as the large subunit of Rubisco (*rbcL*) or the D1 protein of photosystem II (*psbA*). Sequence studies have concentrated for a long time on higher plants and chlorophyll $a+b$-containing algae, thus there were more speculations than data concerning the plastids of chlorophyll $a+c$ algae or red algae. Recent investigations show striking differences between the plastids of different algal taxa. There seems to be no common pattern of gene arrangement and genes which code for plastid proteins found on the nuclear genome in the case of higher plants and green algae such as *rbcS* (which codes for the small Rubisco subunit) are part of the plastid genome of chromophytic and red algae [3, 5, 15, 18, 19, 20]. Trying to interpret those facts phycologists are faced with the question how often the chloroplast-host endosymbiosis took place during evolution. In other words, it is to decide what may be the relationship between different types of plastids and even whether they are of mono- or polyphyletic origin.

In our laboratory we are screening the plastid genomes of several non-chlorophyll $a+b$ algae for the existence of genes which were not found on the completely sequenced genomes of *Nicotiana*, *Oryza* or *Marchantia* [2, 14, 17].

On the plastid genome of two unicellular red algae, *Cyanidium caldarium* (*C. caldarium* Geitler, Allen strain 14-1-1 has been reported to be iden-

The nucleotide sequence data reported will appear in the EMBL, GenBank and DDBJ Nucleotide Sequence Databases under the accession numbers X62578 and X62579.

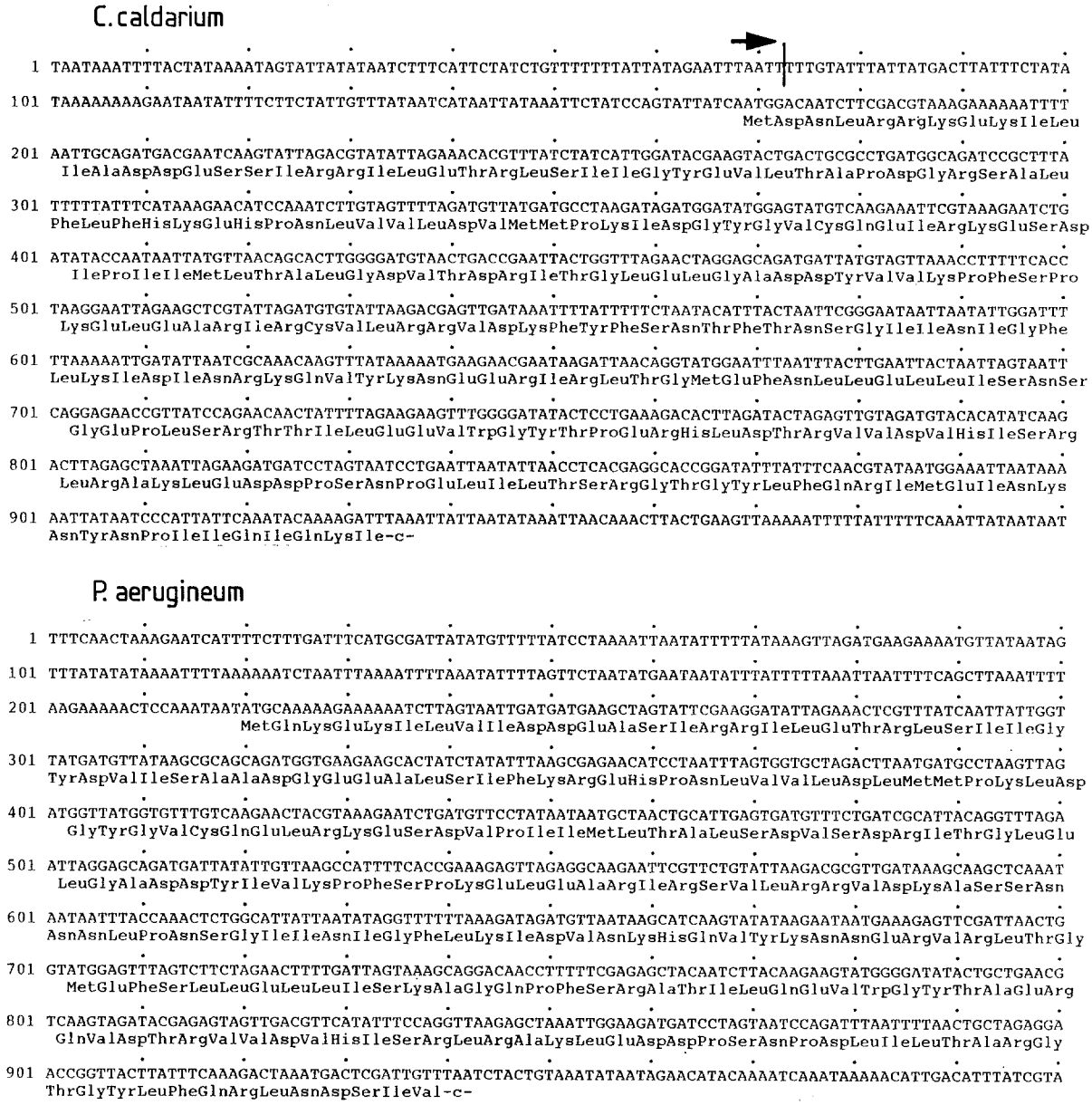

Fig. 1. Nucleotide and amino acid sequences of the two red algal *ompR* homologues. The inverted-repeat/single-copy border in the case of *C. caldarium* is marked by an arrow.

tical with *Galdieria sulphuraria* [13]) and *Porphyridium aerugineum*, we were able to detect an open reading frame (ORF) in the upstream region of the ribosomal operon [7, 8, 9] encoding a protein with 37% (*C. caldarium*) or 40% (*P. aerugineum*) identity and 57% or 58% homology, respectively, to the *ompR* gene product of *Salmonella typhimurium* [6]. The bacterial protein is one part of the two-component regulatory system mediating the osmotic parameters of the procaryote. It has been reported to work as a DNA-binding protein which enables the RNA polymerase to start transcription at less effective promoter motifs [1, 4, 11, 22]. *OmpR*-dependent promoter motifs were

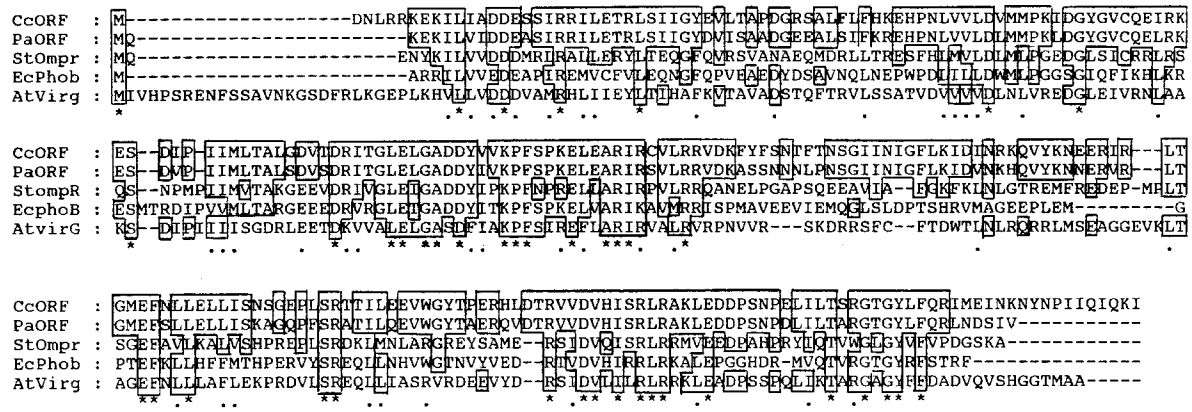

Fig. 2. Amino acid alignment of the two red algal *ompR* homologues with bacterial regulatory proteins. Asterisks mark identical residues, points mark conservative residues. Identical residues in the case of both red algae and other organisms are boxed. Abbreviations: CcORF, *Cyanidium caldarium* open reading frame; PaORF, *Porphyridium aerugineum* open reading frame; StompR, *Salmonella typhimurium* ompR protein; EcphoB, *Escherichia coli* phoB protein; AtvirG, *Agrobacterium tumefaciens* virG protein.

found in the upstream regions of genes of outer membrane-spanning proteins, so-called porins. This raises the question of the possible function of the homologous plastid protein.

There seems to be no homologous gene on chloroplast genomes of higher plants and *Marchantia*, so either there is no need for this regulatory protein or the gene has been transferred to the nucleus. As the amino acid sequence is highly conserved between the bacterial and the rhodoplast protein not only in the N-terminal region which the *ompR* gene product shares with other regulatory proteins such as *phoB* [10] or *virG* [12, 16] but also in the C-terminal part (see Fig. 2), the red algal protein at least should have a homologous function as a promoter-specific protein mediating the interaction between DNA and RNA polymerase. No data are available on red algal plastid RNA polymerase and only few red algal pt genes are sequenced with their promoter regions [5, 18, 19, 20, 21]. Further investigations will have to show whether there are genes homologous to those bacterial porin genes or at least *ompR*-dependent promoters upstream of other rhodoplast genes.

Northern blotting experiments with total cellular RNA failed to detect a transcript but a low level of *ompR*-mRNA might be enough to guarantee the regulatory function of the gene product. Although one may see the plastid *ompR* gene as a relic of the former endosymbiont without any function, the conserved amino acid sequence in the case of two red algae points to a selective pressure on the gene. A *C. caldarium ompR* gene probe hybridizes with the 16S rRNA upstream region of the multicellular red alga *Antithamnion* sp. indicating the existence of the *ompR* homologue on the pt-DNA of a more highly evolved red alga.

Figure 1 shows the nucleotide and amino acid sequences of the two genes and Fig. 2 shows an amino acid alignment of the two red algal *ompR* genes and some bacterial counterparts. Interestingly the *ompR* genes of *C. caldarium* and *P. aerugineum* which are both located close to the genes for the small subunit ribosomal RNA are encoded on different strands. Detailed sequence analyses of the *P. aerugineum* rRNA operon point to a recombination event within this plastid-DNA area in comparison to the red algae *C. caldarium* and *Antithamnion* sp. (manuscript in preparation). It would be of great interest to know more about the existence of *ompR* homologues within the plastid genomes of different taxonomic groups. Further investigations on taxon-specific genes may provide us with phylogenetic markers which will help to elucidate the evolutionary history of the different groups of algae.

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