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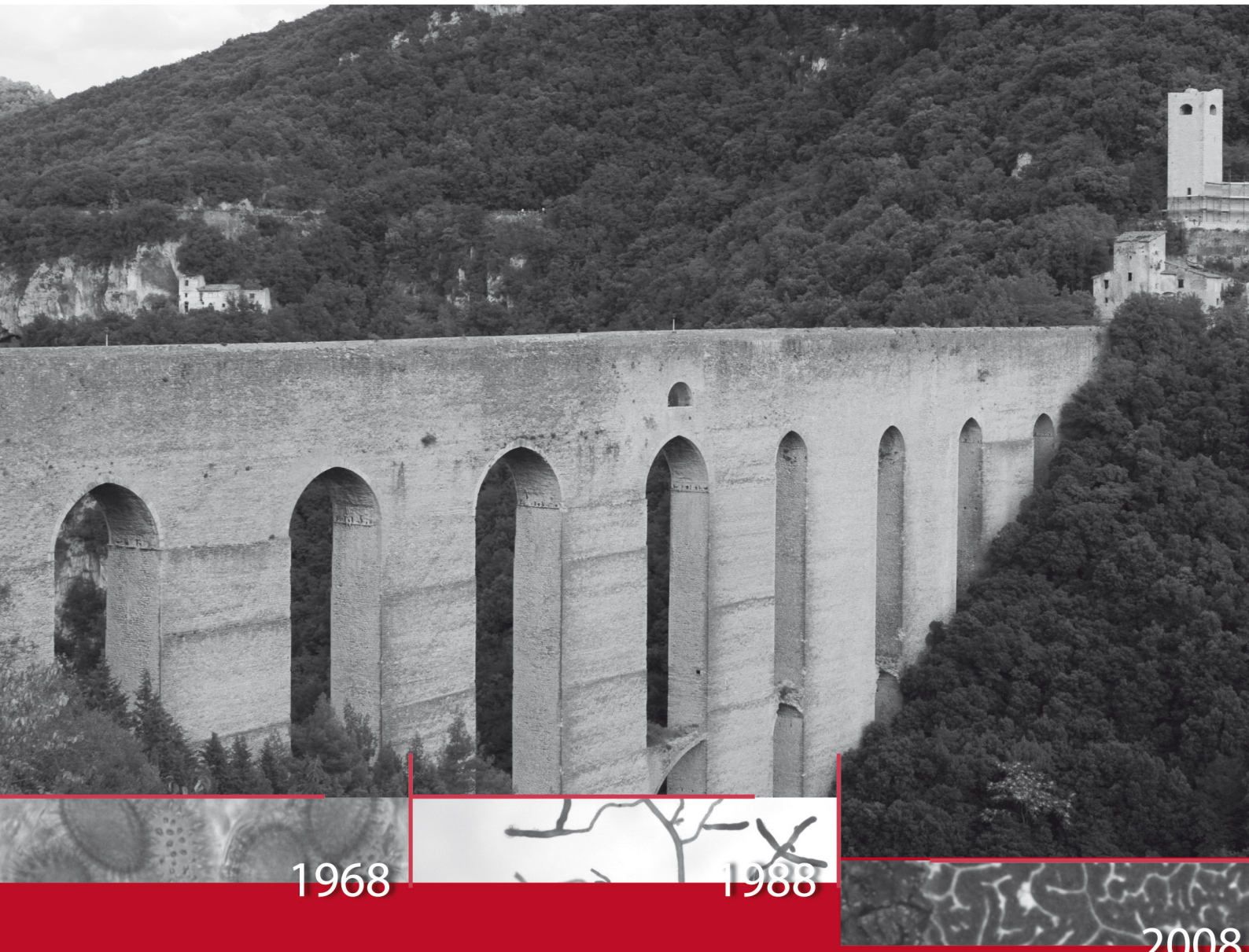

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AN INTEGRATED CELL AND MOLECULAR VIEW POINT OF TRUFFLE BIOLOGY

Balestrini Raffaella, Zampieri Elisa, Abba' Simona, Faccio Antonella, Roberson Robert[1], Bonfante Paola

IPP-CNR and Dipartimento di Biologia vegetale, Viale Mattioli, 25 10125 Torino, Italy;
[1] Arizona State University,Tempe AZ 85287. e-mail: r.balestrini@ipp.cnr.it

Abstract

Specific experiments aimed to assign a biological function to the DNA sequences found in the genome are crucial in a post-annotation phase. In order to archive this aim, mRNAs and gene products localization can be very informative and can confirm the putative function deduced by bioinformatics tools. In the past we have used several cell biology approaches, i.e. in situ hybridization and immunolabelling (immunofluorescence and immunogold) on truffles. These approaches have already permitted the localization of transcripts and several proteins in *Tuber borchii* hyphae during various step of its life cycle. Having the full genome, we suggest that these approaches would be useful to identify the specific roles for genes belonging to a complex gene family. The use of antibodies and the localization of antigens may surely suggest functions when other approaches (i.e. transformants, RNA interference, etc…) cannot be faced. LMD (Laser MicroDissection) is a powerful tool for isolating specific tissues and cell types from sectioned biological specimens, allowing a cell specific extraction of RNA, DNA, or proteins. Preliminary experiments performed on ectomycorrhizae will be presented. We propose the LMD technology to identify genes specifically activated in each of the two fungal compartments (mantle and Hartig net) forming an ectomycorhiza. We have already been applied this technique for gene expression studies in arbuscular mycorrhizae, where the cell specificity for plant and fungal genes involved in phosphate transport has been successfully demonstrated (Balestrini *et al*., 2007 – MPMI 20: 1055–1062). In addition, an accurate analysis at transmission electron microscopy could be also considered a valid support to integrate the molecular analyses, providing information about the presence of specific organelles/structures in fungal hyphae. We are using plunge-freezing procedure followed by freeze substitution to examine and describe the hyphal tip in *Tuber* species. These observations, together with the sequences annotated in the genome and the results derived from the other cell and molecular approaches, could highlight the mechanism at the basis of the hyphal growth in this fungus.

Key words: genomics and cell biology.