

**P024** Membrane-attached hydrogenase I of the hyperthermophilic bacterium *Aquifex aeolicus*  
**Myriam Brugna<sup>a</sup>, Pascale Infossi<sup>a</sup>, Elisabeth Lojou<sup>a</sup>,  
Wolfgang Lubitz<sup>b</sup>, Marianne Guiral<sup>a</sup> and  
Marie-Thérèse Giudici-Ortoni<sup>a</sup>**

<sup>a</sup>Laboratoire de Bioénergétique et Ingénierie des Protéines, CNRS, 31 Chemin Joseph Aiguier, 13402 Marseille cedex 20, France.

<sup>b</sup>Max-Planck-Institut für Bioanorganische Chemie, Stiftstrasse 34-36, D-45470 Mülheim an der Ruhr, Germany.

*Aquifex aeolicus*, a hyperthermophilic bacterium, grows at 85°C under a H<sub>2</sub>/CO<sub>2</sub>/O<sub>2</sub> atmosphere in a medium containing only inorganic compounds. It gains energy for growth from an uncommon electron transfer, from hydrogen to oxygen. In *Aquifex aeolicus*, two [Ni-Fe] hydrogenases are membrane-bound (hydrogenases I and II) and one is cytoplasmic (hydrogenase III). Two of them (hydrogenases I and III) have been purified in the group and a model for the metabolic roles of the three enzymes has been proposed.

Hydrogenase I is composed of 3 subunits, two of them carrying the enzymatic activity, and the third one, a transmembrane dihemic cytochrome *b*, anchoring the enzyme to the membrane, allows the electrons transfer to the quinone in the membrane. This *b*-type cytochrome has been only scarcely studied in bacteria. We have characterized the hydrogenase I of *Aquifex aeolicus*, including the cytochrome *b* subunit, using electrochemistry and EPR spectroscopy. The quinone present in the membrane has been purified and its structure determined using NMR and mass spectrometry. Moreover, isolated hydrogenase I has been incorporated into liposomes containing the quinone of *Aquifex*.