High dark CO2 fixation rates by active chemolithoautotrophic microbes along the water column (100 – 5000 m) off Galicia (NW Iberian margin)

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Previous studies have provided evidences that microbial chemolithoautotrophic activity turns to be substantially larger than hitherto assumed, which could represent a significant source of autochthonous carbon supply in the dark ocean. By using isotopic labeling, we quantified DIC (dissolved inorganic carbon) fixation rates (14C-bicarbonate) and compared it with microbial heterotrophic production (3H-Leucine) in the subsurface, meso- and bathypelagic waters off the northwestern coast of the Iberian Peninsula. Our results revealed that microbial DIC fixation, ranging from 0.05 to 67.07 µmol C m^{-3} d^{-1}, was slightly higher than heterotrophic activity particularly in the meso-pelagic waters. Combining microautoradiography and fluorescence in situ hybridization (MICRO-CARD-FISH), we confirmed that both *Thaumarchaeota* and some bacterial groups, such as SAR 202, SAR 324, SAR 406 and *Alteromona*s actively uptake DIC. Quantitative PCR (QPCR) data revealed that *Thaumarchaeota*, inhabiting the dark waters off the Galician ecosystem, contain the gene for ammonia oxidation and thus are likely chemoautotrophics. QPCR results showed a higher abundance of the thaumarchaeal 16S rRNA and the low ammonia concentration (LAC)-*amoA* ecotype genes in meso- and lower bathypelagic waters than in surface waters. In contrast, high ammonia concentration (HAC)-*amoA* ecotype dominated the subsurface samples. Redundancy analysis (RDA) displayed that environmental and biological variables, such as nitrite and oxygen concentration and the ratio between LAC and HAC-*amo*A gene, correlated significantly with the single-cell identification of active dark bicarbonate fixers and its vertical distribution. Our results provide evidence for the significant contribution to chemolithotrophy by specific archaeal and bacterial groups in the dark ocean.