

Ammonia Oxidising Archaea in the OMZ of a freshwater African Lake

MARC LLIRÓS^{1,2}, ÖZGÜL INCEOGLU³, TAMARA GARCÍA-ARMISEN³, JEAN CRISTOPHE AUGUET⁴, CEDRIC MORANA⁵, FRANÇOIS DARCHAMBEAU⁶, SEAN A CROWE⁷, STEVEN BOUILLON⁵, PIERRE SERVAIS³, ALBERTO V BORGES⁶, JEAN PIERRE DESCY²

¹ Dept Genetics and Microbiology, Universitat Autònoma de Barcelona, Catalunya, Spain

² Laboratory of Freshwater Ecology, University of Namur, Belgium

³ Ecologie des Systèmes Aquatiques, Université Libre de Bruxelles, Belgium

⁴ Equipe Environnement et Microbiologie, Université de Pau et des Pays de l'Adour, France

⁵ Dept Earth and Environmental Sciences, Katholieke Universiteit Leuven, Belgium

⁶ Chemical Oceanography Unit, Université de Liège, Belgium

⁷ Depts Microbiology & Immunology, and Earth, Ocean, & Atmospheric Sciences, University of British Columbia, Canada

Some oceanic or freshwater (stratified lakes) aquatic regions are characterised by the presence of oxygen-deficient water layers (i.e., oxygen minimum zones, OMZ) that represent a minor fraction of total Earth aquatic volume [Paulmier and Ruiz-Pino 2009]. In oceans, OMZ could barely contribute to ca. 50% of bioavailable nitrogen removal [Codispoti et al 2001, Gruber 2008, Lamp et al 2009, Ward et al 2009] supporting diverse microbial communities with critical roles in nitrogen cycling on Earth. Within the nitrification, the ammonia oxidation (i.e., the first step of the reaction) is the rate-limiting stage [Kowalchuk and Stephen 2001]. Until recently, ammonia-oxidizing bacteria (AOB) included in the Beta- and Gammaproteobacteria classes have been considered to be the unique microorganisms responsible for this biochemical process [Purkhold et al 2000]. However, recent evidences highlighted the importance of Archaea and Thaumarchaeota phylum as potential contributors to nitrification [Venter et al 2004, Treusch et al 2005, Könneke et al 2005] especially in oligotrophic marine and freshwater environments [Wuchter et al 2006, Beman et al 2008, Santoro et al 2010]. Lake Kivu is a deep, meromictic and oligotrophic African lake with a permanent chemocline located around 40–60 m depth with a putative community of AOA [Llirós et al 2010, Descy et al 2012]. Two distinct lake water column conditions (i.e., rainy and dry seasons) were analysed in order to detect partitioning of AOA populations by combining 16S rRNA gene pyrosequencing and quantification approaches. Archaea represented a minor fraction of the planktonic microbial community in Lake Kivu (<10%), however AOA represented the main archaeal group in oxic and OMZ water compartments. Besides, no AOB-related sequences were recovered from these water layers and Nitrospira-related sequences were only recovered in the OMZ. Furthermore analyses of the active fraction of the archaeal assemblage revealed AOA as the main active archaeal component in the oxic and OMZ water compartments during rainy but not in dry season. Altogether evidenced the potential implication of AOA and Thaumarchaeota in nitrification processes taking place in the OMZ of Lake Kivu.

REFERENCES Paulmier,A. and Ruiz-Pino,D. 2009. Oxygen minimum zones (OMZs) in the modern ocean. *Progress in Oceanography* 80: 113–128. Codispoti L.A., Brandes J.A., Christensen J.P., Devol A.H., Naqvi S.W.A., Paerl H.W. and Yoshinari T. 2001. The oceanic fixed nitrogen and nitrous oxide budgets: moving targets as we enter the anthropocene? *Scientia Marina* 65, 85–105. Gruber N. 2008. The marine nitrogen cycle: overview and challenges. In: *Nitrogen in the Marine Environment*. (Eds. Bronk D.G., Mulholland D.A., Carpenter M.R. and Edward J.) pp. 1–50. Lam, P., Lavik, G., Jensen, M.M., van de Vossenberg, J., Schmid, M., Woebken, D., et al. 2009. Revising the nitrogen cycle in the Peruvian oxygen minimum zone. *Proc. Natl. Acad. Sci. USA* 106: 4752–4757. Ward, B.B., Devol, A.H., Rich, J.J., Chang, B.X., Bulow, S.E., Naik, H., et al. 2009. Denitrification as the dominant nitrogen loss process in the Arabian Sea. *Nature* 461: 78–81. Kowalchuk, G.A. and Stephen J.R. 2001. Ammonia-oxidizing bacteria: a model for molecular microbial ecology. *Annu. Rev. Microbiol.* 55: 485–529. Purkhold, U., Pommerening-Roser, A., Juretschko, S., Schmid, M.C., Koops, H.P. et al. 2000. Phylogeny

of all recognized species of ammonia oxidizers based on comparative 16S rRNA and amoA sequence analysis: implications for molecular diversity surveys. *Appl. Environ. Microbiol.* 66: 5368–5382. Venter, J.C., Remington, K., Heidelberg, J.F., Halpern, A.L., Rusch, D., Eisen, J.A., et al. 2004. Environmental genome shotgun sequencing of the Sargasso Sea. *Science* 304: 66–74. Treusch, A.H., Leininger, S., Kletzin, A., Schuster, S.C., Klenk, H.-P., and Schleper, C. 2005. Novel genes for nitrite reductase and Amo-related proteins indicate a role of uncultivated mesophilic crenarchaeota in nitrogen cycling. *Environ. Microbiol.* 7: 1985–1995. Könneke, M., Bernhard, A.E., de La Torre, J.R., Walker, C.B., Waterbury, J.B., and Stahl, D.A. 2005. Isolation of an autotrophic ammonia-oxidizing marine archaeon. *Nature* 437: 543–546. Wuchter, C., Abbas, M., Coolen, M.J.L., Herfort, L., van Bleijswijk, J., Timmers, P., et al. 2006. Archaeal nitrification in the ocean. *Proc. Natl. Acad. Sci. USA* 103: 12317–12322. Beman, J.M., Popp, B.N., and Francis, C.A. 2008. Molecular and biogeochemical evidence for ammonia oxidation by marine Crenarchaeota in the Gulf of California. *The ISME Journal* 2: 429–441. Santoro, A.E., Casciotti, K.L., and Francis, C.A. 2010. Activity, abundance and diversity of nitrifying archaea and bacteria in the central California Current. *Environ. Microbiol.* 12: 1989–2006. Llirós, M., Gich, F., Plasencia, A., Auguet, J.-C., Darchambeau, F., Casamayor, E.O., Descy, J.-P., Borrego, C.M. 2010. Vertical distribution of ammonia-oxidizing crenarchaeota and methanogens in the epipelagic waters of Lake Kivu (Rwanda-Democratic Republic of the Congo). *Appl. Environ. Microbiol.* 76: 6853–6863. Descy, J.-P., Darchambeau, F., and Schmid, M. 2012. Lake Kivu. Descy, J.-P., Darchambeau, F., and Schmid, M. (eds) Springer Netherlands.