ESRF	**Experiment title:** Imaging modern and antique microbe-mediated processes in mineralized environments	**Experiment number**: *ME-824*
Beamline: ID22	**Date of experiment**: 10 – 19 June 2004	**Date of report**: December 2005
Shifts: 24	**Local contact(s)**: R. Tucoulou	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

P. Philippot*[1], J. Cauzid*[1,2], J. Foriel*[1], B. Ménez*[1], C. Rommevaux*[1], N. Thébaud

[1] Laboratoire Géobiosphère Actuelle et Primitive, IPGP Paris, case 89, 4 place Jussieu, 75252 Paris 05

[2] ESRF, line ID22, ID21

ESRF	**Experiment title:** Imaging modern and antique microbe-mediated processes in mineralized environments	**Experiment number**: *ME-824*
Beamline: ID22	**Date of experiment**: 25 April to 3 May 2005	**Date of report**: December 2005
Shifts: 21	**Local contact(s)**: P. Bleuet, Gema MARTINEZ CRIADO	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

P. Philippot*[1], J. Cauzid*[1,2], E. Gérard*[1], K. Lepot*[1] B. Ménez*[1], C. Rommevaux*[1], N. Thébaud

[1] Laboratoire Géobiosphère Actuelle et Primitive, IPGP Paris, case 89, 4 place Jussieu, 75252 Paris 05

ESRF	**Experiment title:** Imaging modern and antique microbe-mediated processes in mineralized environments	**Experiment number**: *ME-824*
Beamline: ID22	**Date of experiment**: 5 to 13 December 2005	**Date of report**: December 2005
Shifts: 21	**Local contact(s)**: P. Bleuet, Gema MARTINEZ CRIADO	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

P. Philippot*[1], J. Cauzid*[1,2], E. Gérard*[1], K. Lepot*[1] B. Ménez*[1], C. Rommevaux*[1]

[1] Laboratoire Géobiosphère Actuelle et Primitive, IPGP Paris, case 89, 4 place Jussieu, 75252 Paris 05

[2] ESRF, line ID22, ID21[1] Laboratoire Géobiosphère Actuelle et Primitive, IPGP Paris,

The aim of this Long Term Project was to bring together a variety of specialists of different scientific horizons to image and quantify living and fossil biomaterial in mineralized environment and fluid inclusions using a variety of tracers such as sulfur, lipids, selenium, arsenic and iron. By exploring the full spectrum of microbial ecology in extreme environments, this has provided important new insights into life-sustaining processes in the rock record and contribute to the understanding of the environmental conditions and the origin of life on the Primitive Earth.

We have obtained a total of 44 shifts on ID21 and 82 shifts on ID 22 from April 2004 to December 2005.

On ID 22, the 82 shifts were dedicated to:

I) Fluid inclusion studies: Development and applications. This study is part of the PhD of Jean Cauzid who was supported by a CNRS-ESRF grant current 2002-2005. Jean Cauzid has been hired as a postdoctorate on ID22 this year for a tree year period starting January 2006

II) 3D imaging of microorganisms in mineralized samples

III) Intracellular XANES spectroscopy at iron K edge on frozen hydrated neuron cells

I – Fluid inclusion analysis – Jean Cauzid PhD thesis (ESRF – CNRS grant) and applications to hydrothermal systems from Archaean environment (Foriel and Thébaud PhD Thesis; 10-19 June 2004; 6 shifts of november 3-13, 2004; 25 April – 3 May 2005; and 9 shifts of December 5-13, 2005).

Fluid inclusions are small quantities of fluids trapped in minerals. A single inclusion traps a few cubic micrometers of fluid, which migrates in km^3 volumes through the Earth's crust. Variability between inclusions must therefore be smoothed out to allow extraction of information about past fluid-rock interactions from such samples. This is fulfilled through analysis of statistically relevant

series of fluid inclusions. The quantification algorithm has therefore been computed to facilitate processing of fluorescence data measured on numerous single fluid inclusions. Quantification of low Z elements (S to Ca) was made possible by the use of the He chamber developed during the previous LTP (ME401). This fluorescence-based quantification procedure was improved with combining fluorescence and transmission measurements (Cauzid et al., 2004). A standardless quantification procedure was simultaneously established and enhanced by statistical error calculations (Cauzid et al., in press).

This method for analysing homogeneous fluid inclusions was applied to two studies. The first one aimed at measuring the concentration of halogens (Cl, Br) and sulphate in Archaean seawater preserved as fluid inclusions in quartz crystals. Knowledge of these concentrations has important implications for understanding the evolution of Earth's hydrosphere and atmosphere and the development of early life. Their analysis provided evidences of life as early as 3.8 billion years ago in shallow marine, closed basin environments (Foriel et al., 2004) and published as a ESRF Highlights in 2004 (Philippot et al., 2004). A second application focused on fluids associated with Archaean gold mineralization (Thébaud et al., 2006 a,b, in review).

Fluid inclusions created at depth in the Earth's crust seal homogeneous fluids. However, they often nucleate vapour, immiscible liquid phases and daughter minerals upon cooling, hence become highly heterogeneous. They can be homogenised again by heating to the temperature at which they were created (Figure 1). Two tracks were followed for quantification of such fluid inclusions: punctual fluorescence measurements on fluid inclusions homogenised in situ by heating, and quantification at room temperature of heterogeneous inclusion through pre-treatment of 2D fluorescence imaging data. Both delivered comparable results showing that quantification of heterogeneous closed systems is possible (Cauzid et al., submitted to Geochim. Cosmochim. Acta).

These developments enabled analysis of naturally co-generated liquid and vapour inclusions from room temperature to 500°C. Measurements combined punctual micro-fluorescence, 2D micro-fluorescence imaging (Figure 2) and micro-XANES (Figure 3). It aimed at understanding chemical processes explaining Cu partitioning into the vapour during liquid-vapour immiscibility in the lithosphere. 2D fluorescence imaging of Cu and Fe disproves results obtained by another research team. A helical fluorescence tomography gave an external check on Fe and Cu distributions. 3D elemental distributions match our 2D imaging experiments (Figure 4; Cauzid et al., submitted to NIM B) and the quantitative side of fluorescence tomography supports our quantification results obtained on heterogeneous fluid inclusions. Results show that Cu enrichment in the vapour is due to complexion with S, but that it evolves toward Cl-ligand complexes upon cooling. These results give information on the genesis of porphyry and epithermal ore deposits, which are major Cu resources. (Cauzid et al., submitted to Geochim. Cosmochim. Acta).

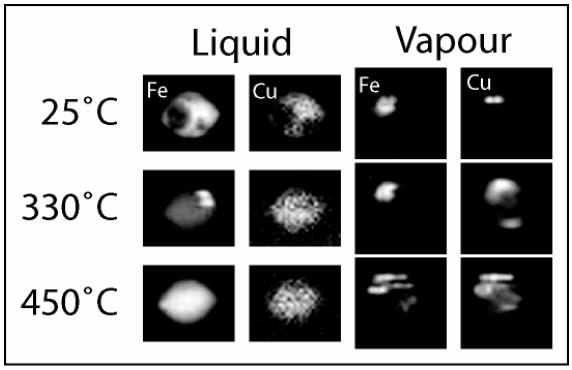

Figure 1: Fe and Cu homogenisation in vapour and liquid inclusions with heating.

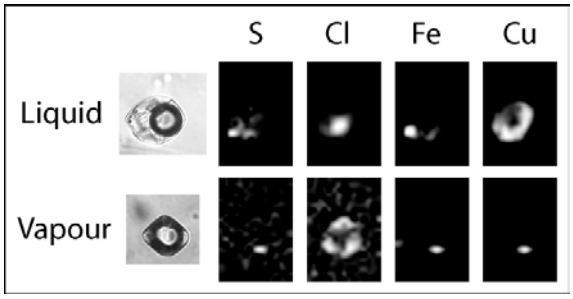

Figure 2: Optical view and 2D maps of S, Cl, Fe and Cu in individual liquid (top) and vapour (bottom) inclusions. Optical microphotograph and corresponding fluorescence maps do not coincide because the first one is taken normal to the wafer surface whereas maps are obtained using a fluorescent beam emerging at 45° from the sample surface.

Figure 3: XANES spectra measured at the Cu K_α edge in vapour and liquid inclusions as a function of temperature. The 506°C XANES spectrum is missing in the liquid inclusion due its decrepitation in the 458-506°C temperature range.

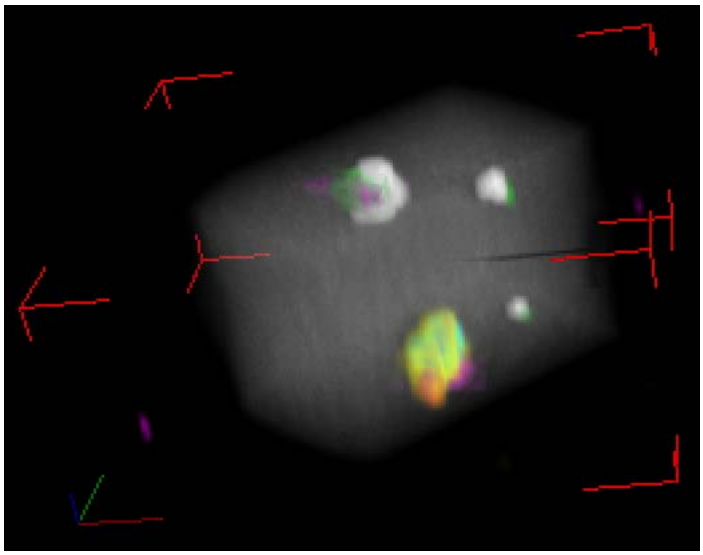

Figure 4: 3D reconstruction of transmission (white), Si(grey), Fe (red), Cu (green), Zn (blue), As (violet) and Br (yellow) in a quartz sample hosting one liquid and three vapour inclusions. Vapour inclusions are mainly recognised through the transmitted signal (white-dominated zones); the liquid inclusion is recognised with the Br signal (yellow dominated zone).

REFERENCES

Cauzid, J., Philippot, P., Bleuet, P., Simionovici, A., Golosio, B. et Somogyi, A., 3D imaging of vapour and liquid fluid inclusions from the Yankee Lode deposit, Mole Granite, Australia using helical fluorescence tomography. Nuclear Instruments and Methods in Physics Research: Section B: submited

Cauzid, J., Philippot, P., Martinez-Criado, G., Ménez, B. et Labouré, S., Contrasting Cu-complexing behaviour in vapour and liquid fluid inclusions from the Yankee Lode deposit, Mole Granite, Australia. American Mineralogist: submitted.

Cauzid, J., Philippot, P., Somogyi, A., Ménez, B., Simionovici, A. et Bleuet, P., Standardless quantification of single fluid inclusions using synchrotron radiation induced X-ray fluorescence. Chemical Geology, in press.

Cauzid, J., Philippot, P., Somogyi, A., Simionovici, A. et Bleuet, P., 2004. Quantification of single fluid inclusions by combining Synchrotron Radiation-Induced μ-X-ray Fluorescence and Transmission. Analytical Chemistry, 76: 3988-3994.

Foriel, J., Philippot, P., Rey, P., Somogyi, A., Banks, D.A. et Ménez, B., 2004. Biological control of Cl/Br and low sulfate concentration in a 3.5-Gyr-old seawater from North Pole, Western Australia. Earth and Planetary Science Letters, 228: 451– 463.

Philippot, P., Cauzid, J., Foriel, J., Somogyi, A., Ménez B. and Rey, P., 2005. Modern Composition of 3,500 Million year- old Seawater: Implications for Life on the Primitive Earth, ESRF Highlights 2004, 111-112

Thébaud N, Philippot P, Rey P, Brugger J, Van Kranendonk M, Grassineau N, 2006. Protracted fluid-rock interaction in the mid-Archaean and implication for gold mineralization: Example from the Warrawoona Syncline (WA), Earth Planet Sci Lett., in review

Thébaud N, Philippot P, Rey P, Cauzid, J. 2006. Composition and origin of fluids associated with lode gold deposits in a mid Archaean greenstone belt (Warrawoona Syncline, Pilbara Craton, Western Australia) using Synchrotron Radiation X-Ray Fluorescence. Contrib. Mineral. Petrol., in review

+ 18 abstracts presented at international meetings among which 3 as invited speakers (American Geophysical Union, San Francisco, December 2004; International Mineralogical Association, Kobe, July 2006; Goldschmidt Conference, Melbourne August 2006).

II- 3D imaging of microorganisms in mineralized samples (12 shifts of 5-13 décembre 2005)

During these last two years, we had planned to go further in the in situ experimentation of microbial activity. Accordingly; the preliminary results obtained previously had received a great deal of interest in the geochemical and microbiological communities. However to improve the measurements, the development of a more adapted microreactor was required which was not brought to a successful conclusion for strategical and technical reasons. Considering the very promising results obtained on beamline ID21 on the 2D imaging of microorganisms in mineralized samples, we evaluated the possibility of associating scanning X-ray microscopy available on ID22 to the protocol based on fluorescent in situ hybridization coupled to ultra-small immunogold detection and described above. However, owing to the higher X-ray energies used on this beamline, the spatial resolution was not adapted and we were not able to image individual cells. In a second step, to take advantage of the low absorption of hard X-rays available on ID22, we developped a new analytical protocol allowing to recognize, on the basis of immunodetection, various molecular components of natural biofilms. Such resistance structure are indeed, the main survival strategy of microbial communities in the deep subsurface. We aim at localizing biofilms within rock fractures by combining X-ray tomography and fluorescence tomography together with metallic particles associated with specific proteins or antibodies and displaying higher density contrast or fluorescence yield compared to the host matrix. This will allow 3D localization of microbial biofilms present within rocks sample at the core scale. However, we only had beamtime for a preliminary study that do not provide sufficient fruitfull results. and dedicated proposals will be submitted in the upcoming months to obtain beamtime for this purpose.