	Experiment title: Structural studies of human HDAC8 in complex with a peptidic substrate and an active site inhibitor	**Experiment number**: MX-394
Beamline: ID14 4 ID14 3	**Date of experiment**: from: 16 May 2005 to: 17 May 2005 from: 18 June 2005 to: 20 June 2005	**Date of report**: 24-July-2005
Shifts: 3 6	**Local contact(s)**: Xavier Thibault Joanna Timmins	*Received at ESRF:*

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Report:

Histone deacetylases (HDACs) are enzymes that remove acetyl groups from lysines near the ends of the histones, the proteins wrapped in the DNA of chromosomes. The removal of the acetyl groups, or deacetylation, causes chromatin condensation, which leads to transcriptional repression. HDACs play an important role in controlling gene transcription but little is known about how they work at a molecular level (how they catalyze the deacetylation reaction, how they are regulated, how they interact with the histones, etc). The aberrant recruitment of HDACs has been mechanistically linked to malignancy in leukemias and lymphomas. HDACs also play fundamental roles in cell differentiation and hypertrophy, cell cycle progression, mitosis, genome stability and stress responses, which are all processes altered frequently in cancer. In line with the biological roles of HDACs, small molecule HDAC inhibitors show antitumor activity in preclinical models and in clinical trials and are promising to become effective, new antineoplastic therapeutics. We have published recently the crystal structure of the first human HDAC, HDAC8, [Vannini *et al., Proc. Natl. Acad. Sci. U S A*, 101 (42), 15064-15069 (2004)], using data collected at ESRF. Knowing the three-dimensional structure of HDAC8 now allows us to start to understand some of the details of how HDAC enzymes work. Our structure has suggested several mutations for further structural work. We have

produced and crystallized several mutants to obtain crystal structures of HDAC8 in complex with substrates and with new inhibitors of different classes. We have collected very recently five data sets:

1. HDAC point mutant in complex with a peptidic substrate. Space group P21, cell dimension 51, 130 56 Å, β 116°. Resolution 2.0 Å, Completeness 95.6 %, Rmerge 4.9 %
2. HDAC8 point mutant in complex with the same peptidic substrate as point 1, but of a different space group. Space group P212121, cell dimensions 82, 98, 105 Å. Resolution 2.6 Å, Completeness 87.7 %, Rmerge 10.6 %
3. HDAC8 double mutant in complex with the same peptidic substrate as point 1. Space group P212121, cell dimensions 81, 91.89, 193.35 Å. Resolution 2.85 Å, Completeness 80 %, Rmerge 10.3 %
4. HDAC8 point mutant in complex with an hydroxamic acid compound. Space group P212121, cell dimensions 84, 98, 110 Å. Resolution 2.5 Å, Completeness 100 %, Rmerge 10.3 %
5. HDAC8 point mutant in complex with the same hydroxamic acid compound as point 4. Space group P212121, cell dimensions 84, 98, 110 Å. Resolution 2.0 Å, Completeness 98.7 %, Rmerge 8.6 %

Structure refinement/Model building of these structures are in progress.

In addition we have other crystals with different compounds/peptides and co-crystallization experiments of several other mutants are in progress. We would like to measure the already available crystals plus any new crystals during the next proposal (MX-11327).