ROBL-CRG	**Experiment title: Binding forms of actinides (U(VI)) and lanthanides (Eu(III)) with single constituents of biofluids (saliva, urine)**	**Experiment number**: 20-01-685
Beamline: BM 20	**Date of experiment**: from: 11/02/09 to: 14/02/09	**Date of report**: 13/08/09
Shifts: 9	**Local contact(s)**: C. Hennig	*Received at ROBL:*

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Report: Synchrotron-based EXAFS is a powerful technique to obtain structural information on radionuclide bioligand species. Within this proposal we explore structural parameter of U(VI) and Eu(III) species with the protein α-amylase (Amy). This enzyme is to be found in biofluids like saliva and pancreatic fluid and is responsible for starch degradation.

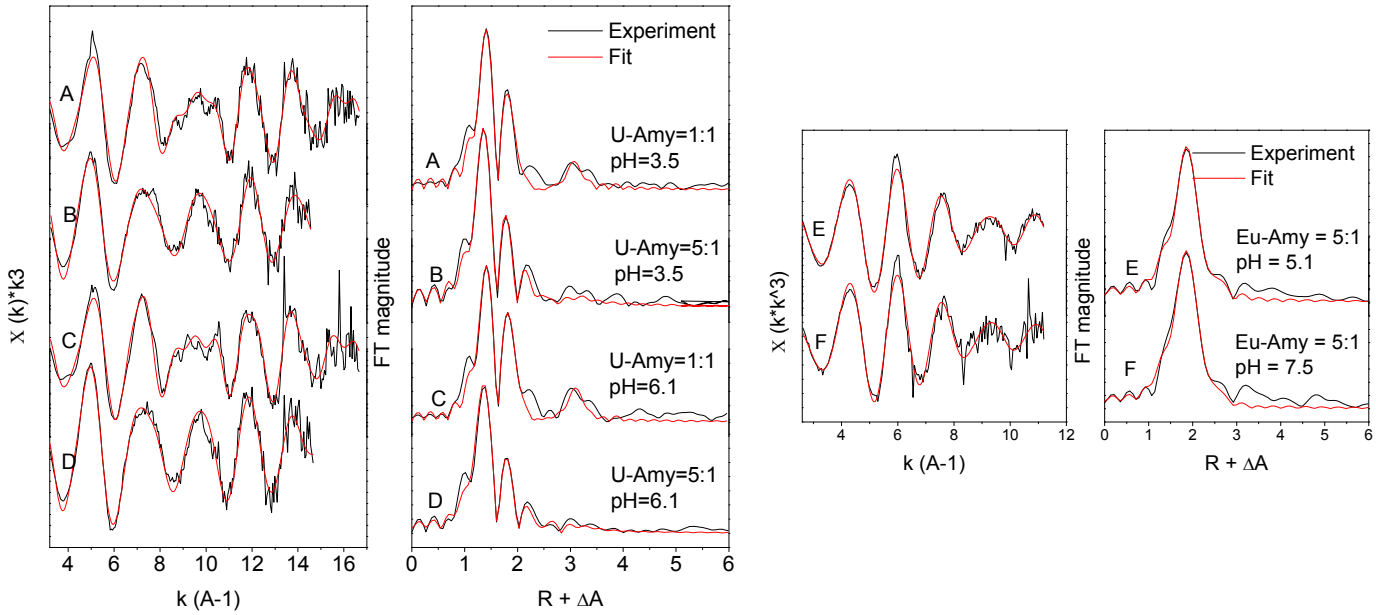

Fig. 1. U L_{III} edge (left) and Eu L_{III} (right) k^3-weighted EXAFS spectra and the corresponding Fourier transforms and the theoretical fits (red lines).

Experimental. U L_{III} and Eu L_{III} edge EXAFS measurements were carried out with wet pastes of α-amylase after contact with concentrated UO_2^{2+} or Eu^{3+} solutions, adjusted to a fixed pH (see Table 1). These samples were measured at 15 K (U(VI)) and room temperature (Eu(III)) either in transmission or in fluorescence mode.

Results. A selection of the measured EXAFS oscillations and corresponding Fourier transforms are presented in Fig. 1. The extracted structural parameters are summarized in Table 1.

Table 1. Summary of the determined structural parameters.

Sample	**Shell**	**N**	**R [Å]**	**σ^2 [Å^2]**	**ΔE_0 (eV)**
A: 1.8 µmol UO_2^{2+}, 100 mg Amy, pH = 3.6	U=O U-O_{eq} U-C	2* 3.1(2) /3.1	1.79 2.28(1) 3.47(1)	0.00124 0.00276 0.00153	5.4
B: 9.0 µmol UO_2^{2+}, 100 mg Amy, pH = 3.6	U=O U-O_{eq1} U-O_{eq2} U-C	2* 2.8(8) 1.9(4) /1.0	1.78 2.30(2) 2.48(2) 2.84(2)	0.00164 0.004078 0.004148 0.002f	4.8
C: 1.8 µmol UO_2^{2+}, 100 mg Amy, pH = 6.1	U=O U-O_{eq} U-C	2* 3.4(3) /3.4	1.79 2.28(1) 3.48(1)	0.0015 0.00244 0.00202	5.1
D: 9.0 µmol UO_2^{2+}, 100 mg Amy, pH = 6.1	U=O U-O_{eq1} U-O_{eq2} U-C	2* 3.1(6) 1.1(6) /0.6	1.79 2.33(1) 2.52(2) 2.93(2)	0.00208 0.005056 0.001257 0.00603	5.2
E: 13 µmol Eu^{3+}, 500 mg Amy, pH = 5.1	Eu-O Eu-C1 Eu-C2	10.3(4) 2.7(6) 4*	2.41(1) 2.96(1) 3.42(1)	0.00984 0.00177 0.00945	-11.5
F: 13 µmol Eu^{3+}, 500 mg Amy, pH = 7.5	Eu-O Eu-C1 Eu-C2	10.8(7) 4.8(1,6) 2*	2.39(1) 2.95(1) 3.41(3)	0.0103 0.00614 0.0049	-13.2

** fixed parameter, / linked parameter.*

The uranyl amylase samples with similar composition and only different pH are very similar among each other. The coordination sphere in the uranyl equatorial plane seems to be independent from the pH but only dependent from the uranyl to protein ratio. The short U-Oeq distances, connected with a quite small coordination number for the samples with lower uranyl concentration (A and C) can be interpreted as a four-fold coordination with carbonyl groups. The splitted equatorial shell for the samples with higher uranyl concentration (B and D) can be interpreted as a mixed coordination with monodentate carbonyl groups (shorter U-O_{eq1} distances) and bidentate carboxyl groups (longer U-O_{eq2} distances).

The EXAFS spectra of Eu(III) amylase samples are also quite similar among each other at different pH. It seems to be a mixed coordination with carbonyl and carboxyl groups and some remaining water molecules.

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