Interaction of Eu(III) and Cm(III) with mucin – A key component of the human mucosa

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To evaluate the potential health risks caused by the ingestion of lanthanides (Ln) and actinides (An), investigations into the chemical behavior of these metals in the human gastrointestinal tract are necessary. Mucin is an important part of the protective mucosa layer in the digestive system. We have recently reported that mucin interacts strongly with Eu(III) and Cm(III), representatives of Ln(III) and An(III), respectively, under *in vivo* conditions.[1] In order to investigate the complexation behavior of this protein with Ln(III)/An(III), TRLFS measurements were performed on Eu(III)/Cm(III)-mucin solutions with different protein concentrations and at different pH. The results indicate the formation of at least two independent mucin species. At higher pH, the formation of hydroxide species was also observed.

In general, the lanthanide and actinide elements have no significant vital roles. However, through different processes such as nuclear accidents, these heavy metals could be potentially released into the environment where they could be incorporated into the food chain and eventually into the human body through oral ingestion. Because of the chemical- and radiotoxicity of An, it is important to understand their chemical and biological behavior under *in vivo* conditions for the reliable health risk assessment of these elements. Our previous report has revealed that the protein mucin is an important binding partner for Eu(III) and Cm(III) in the human gastrointestinal tract.[1] Mucins are high molecular weight glycoproteins and an important component of mucosa, which is a thick and viscoelastic layer covering the whole digestive system. Mucosa also acts as a protective barrier to pathogens and toxic substances. Responsible for its protective functions is the protein mucin.[2] Based on this background, this study focuses on the interaction of this protein with Eu(III) and Cm(III) as representatives of the Ln(III) and An(III), respectively.

EXPERIMENTAL. A solid form of mucin was weighed and dissolved in an aqueous solution to mix with Eu(III) or Cm(III). The final metal concentrations in the sample solutions were 1×10^{-5} M and 3×10^{-7} M for Eu(III) and Cm(III), respectively. For the constant pH experiments, the mucin concentration was ranged from 0.01 to 2 mg/mL, while the pH was varied between 1 and 10 with a mucin concentration of 1 mg/mL for the constant mucin concentration experiments. All sample solutions had a constant ionic strength of 0.1 M NaCl. The TRLFS measurements of the prepared samples were carried out at room temperature. For radiation safety reasons, the measurements with Cm(III) were performed in a glove box under nitrogen atmosphere, while the measurements with Eu(III) were carried out in ambient atmosphere.

RESULTS. In Fig. 1, the luminescence spectra of Eu(III) (left) and Cm(III) (right) with different mucin concentrations are shown. The Eu(III) spectra exhibit an increasing trend in luminescence intensity associated with the $^5D_0 \rightarrow {}^7F_2$ emission band at ~615 nm with increasing the mucin concentration. Additionally, the presence of the symmetry forbidden $^5D_0 \rightarrow {}^7F_0$ emission band at ~580 nm indicates the formation of asymmetric complexes. These results suggest the complex formation between Eu(III) and mucin. The Cm(III) spectra show a red shift with increasing mucin concentration and the spectral shape is unchanged above the mucin concentration of 0.3 mg/mL. At the highest mucin concentrations, at least two independent species were detected with the average luminescence lifetimes of $\tau_1 = 146 \pm 7$ µs and $\tau_2 = 431 \pm 16$ µs, and $\tau_1 = 94 \pm 3$ µs and $\tau_2 = 268 \pm 5$ µs for Eu(III) and Cm(III), respectively. The longer lifetimes correspond to 6–7 water molecules in the first coordination sphere, while the shorter lifetimes suggest ~2 water molecules remaining in the first coordination sphere.

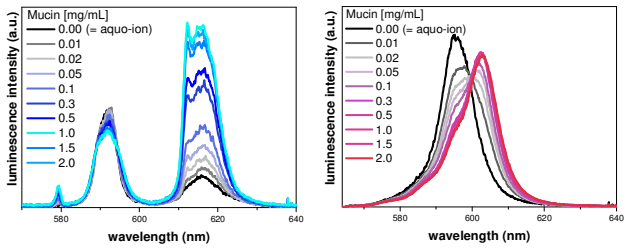

Fig. 1: Luminescence spectra of Eu(III) (1×10^{-5} M, left) and Cm(III) (3×10^{-7} M, right) with different mucin concentrations at pH 4.5 and room temperature.

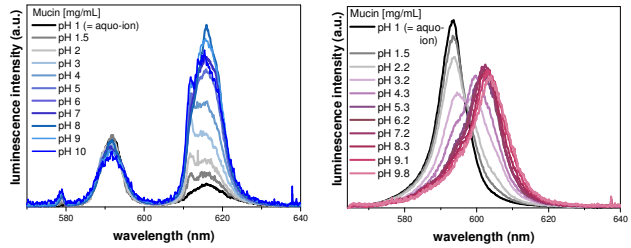

Fig. 2: Luminescence spectra of Eu(III) (1×10^{-5} M, left) and Cm(III) (3×10^{-7} M, right) at different pH with a constant mucin concentration of 1 mg/mL at room temperature.

As shown in Fig. 2, the luminescence spectra of Eu(III) at different pH show again an increasing trend in the luminescence intensity associated with the $^5D_0 \rightarrow {}^7F_2$ emission with increasing the pH up to 8. However, the luminescence intensity decreases at pH 10 due to the formation of hydroxide species. The Cm(III) spectra at different pH indicate a red shift with increasing pH and a peak splitting at pH 3.2. As observed for the Eu(III) results, there is a decrease in the Cm(III) luminescence intensity at pH 10 because of the formation of hydroxide species. The luminescence lifetimes decay bi-exponentially for both experiment series. At pH 8–9, which represents the highest pH range before the hydroxide precipitation occurs, the average lifetimes are $\tau_1 = 362 \pm 15$ µs and $\tau_2 = 784 \pm 22$ µs, and $\tau_1 = 131 \pm 7$ µs and $\tau_2 = 403 \pm 12$ µs for Eu(III) and Cm(III), respectively. These lifetimes correspond to 2–4 and 1 water molecules in the first coordination sphere.

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[1] Wilke, C. *et al.* (2015) *Report HZDR-059*, p.12.
[2] Bansil, R. *et al.* (2006) *Curr. Opin. Colloid Interface Sci.* **11**, 164–170.