

## Development of $^{67}\text{Cu}$ -labeled somatostatin derivatives to optimize nuclear medicine therapy

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Nuclear medicine therapy has garnered significant attention in recent years as an effective treatment process that places less burden on patients. Of the various nuclides used for this type of treatment, four are available in Japan, namely,  $^{131}\text{I}$ ,  $^{90}\text{Y}$ , and  $^{177}\text{Lu}$ , which are  $\beta$ -particle-emitting nuclides, and  $^{223}\text{Ra}$ , which is an  $\alpha$ -particle-emitting nuclide.  $^{90}\text{Y}$  emits high-energy  $\beta$  particles with energies of 2.28 MeV and exhibits good therapeutic effect; however, it has a significant impact on the surrounding tissues, requiring the injected dose to be limited to minimize exposure of the other organs to the radiation.<sup>1)</sup>

Several other nuclides can potentially be used for nuclear medicine therapy. For example,  $^{67}\text{Cu}$  emits low-energy  $\beta$  particles with energies of 0.392 MeV; as such, it can be administered in large doses, but its effectiveness has not yet been sufficiently analyzed. Although the range of its therapeutic effect is limited owing to its low energy, good results may be obtained with  $^{67}\text{Cu}$  for small tumors, thus leading to effective and efficient treatment.<sup>2)</sup> We previously synthesized  $^{67}\text{Cu}$ -ToDBTTATE as a peptide derivative targeting the somatostatin receptor and demonstrated that this compound constricts tumor growth in model mice bearing AR42J rat pancreatic tumor cells.<sup>3)</sup>

Meanwhile,  $^{177}\text{Lu}$ -labeled octreotate, which also targets the somatostatin receptor, is already being used clinically for the treatment of neuroendocrine tumors. However, this radiopharmaceutical can be problematic in that the kidney is easily exposed to its radiation. Thus, to reduce renal radioactivity, an amino acid preparation is administered via an intravenous drip infusion before the administration of  $^{177}\text{Lu}$ -octreotate. Another method to reduce accumulation in the kidney, as reported by Suzuki *et al.*, is to use a tripeptide substrate that is rapidly degraded by the brush border membrane (BBM) enzymes present on the lumen of the renal tubule.<sup>4)</sup>

In this study, we developed a new compound that incorporates a tripeptide linkage that is rapidly degraded by the enzymes on the renal BBM into its molecule and investigated its distribution in mice.

First, the compound was synthesized and labeled with  $^{67}\text{Cu}$ . Then, the labeled compound was incubated for 1 hour at  $37^\circ\text{C}$  in BBM vesicles, and the

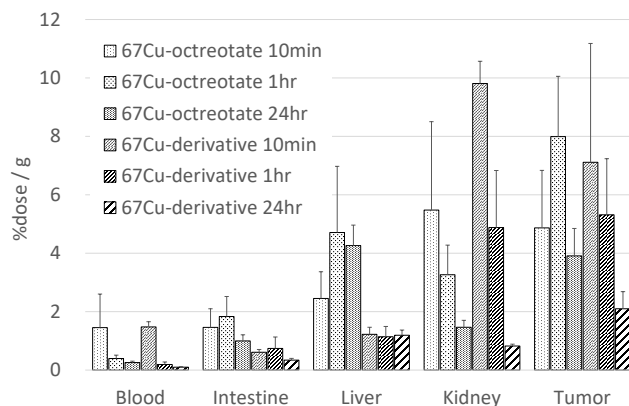


Fig. 1. Comparative biodistribution of radioactivity after intravenous injection of  $^{67}\text{Cu}$ -labeled derivatives in mice ( $n = 4$ ).

radiolabeled species in the reaction mixture were analyzed via RP-TLC to estimate renal BBM-mediated cleavage of the peptide linkage. BBM vesicles were isolated from the renal cortex of male Wistar rats using the Mg/EDTA precipitation method, as had been reported previously.<sup>5)</sup> For the biodistribution studies, tumor-bearing mice were prepared by implanting AR42J tumor cells ( $5 \times 10^6$  cells) in 0.1 mL PBS into the flanks of nude mice (BALB/c-nu/nu, male). Once the tumors had grown, the  $^{67}\text{Cu}$ -labeled compounds were administered intravenously. The mice were killed at 10 min, 1 hour and 24 hours after administration, and tissues of interest were excised and weighed before their radioactivities were measured. The results are presented as a percentage of the injected dose per gram of tissue weight. The animal studies were conducted in accordance with the Guide for the Care and Use of Laboratory Animals of Suzuka University of Medical Science.

The  $^{67}\text{Cu}$ -labeled somatostatin derivatives were obtained with high radiochemical purities ( $>95\%$ ). After 1 hour of incubation with BBM vesicles, a  $^{67}\text{Cu}$ -labeled derivative with a tripeptide linkage generated some radiometabolites. The biodistribution study in mice showed that the initial kidney accumulation of this derivative increased slightly, but also that the clearance from the kidney was faster than that of  $^{67}\text{Cu}$ -labeled octreotate. In the liver, this derivative exhibited consistently lower accumulation levels across all time points, which may be related to its rapid clearance from the kidney.

The  $^{67}\text{Cu}$ -labeled somatostatin derivative with a tripeptide linkage exhibited high elimination rates of radioactivity from the kidney and reduced the renal

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localization of the radiopharmaceutical. In the future, we plan to further optimize the molecular structures of the derivatives and create useful compounds.

#### References

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