学 位 論 文 内 容 の 要 旨

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学 位 論 文 題 名

Basis research and development of DNA-targeted radio-platinum agents
for Auger electron cancer therapy
（Auger 電子を用いたがん治療創成に向けた
DNA を標的とする放射性白金薬剤の基礎開発研究）

In targeted radionuclide therapy (TRT), β^--rays are the most commonly used in the clinic, exhibiting cytotoxicity for widespread cancer cells because of their emission in a moderate range (0.05–12 mm) of energetic electrons. However, recently, α-rays have attracted a great deal of interest because of their high therapeutic efficacy. α-rays have a high linear energy transfer (LET) (80 keV/μm) in a short range (2–500 nm), minimizing extra doses to normal cells compared to β^--rays. Auger electrons are candidate sources in TRT because of a high LET (4–26 keV/μm) in a nano-scale range (2–500 nm). Auger electrons potentially cause the least amount of damage to predominately the targeted molecules and cancer cells, meaning an ideal source in internal radiotherapy without any adverse effects.

The distance between DNA and an Auger electron emitter is expected to be crucial when inducing fatal damage by Auger electrons for therapy. Agents should bind to DNA as close as possible; however, such DNA-targeting radiopharmaceuticals capable of binding to DNA efficiently and damaging DNA effectively with Auger electrons have yet to be established. Auger electron-emitting radio-Pt is a promising candidate in DNA targeting because Pt itself binds to DNA directly. Here, in the present thesis, basis research and development of DNA-targeted radio-platinum agents were conducted for future Auger electron cancer therapy.

Chapter 1: Development of a Production Method for No-Carrier-Added Radio-platinum and DNA Damage Evaluation for Auger electrons using Radio-cisplatin

In order to confirm the potential of radio-Pt for DNA-targeted Auger electron therapy, it is necessary to investigate the degree of the DNA-damaging effect of Auger electrons released from radio-Pt that binds to DNA. In *chapter 1*, at first, a novel method for no-carrier-added (n.c.a.) radio-Pt production from an Ir target using accelerators was developed. N.c.a. radio-$Pt^{II}Cl_4^{2-}$ was successfully prepared in a useful chemical form as a precursor for the synthesis of platinum drugs. Thereafter, the DNA damaging effect of Auger electrons was evaluated using n.c.a. radio-cisplatin with the negligible

chemical effect of nonradioactive Pt. The observations of the DNA damage by n.c.a. radio-cisplatin indicated that double-strand breaks (DSBs) were induced by Auger electrons released from radio-Pt bound to DNA, not the chemical effect of nonradioactive Pt carriers. Unfortunately, due to its low cellular uptake, only a small fraction of radio-cisplatin bound to DNA, leading to a low probability of DSBs and low cytotoxicity. To maximize the damaging effect of Auger electrons and reveal their ideal potential, it was necessary to develop a more efficient system for delivering radio-Pt to DNA.

Chapter 2: Development of Radio-Pt Labeled Agents Binding to DNA Efficiently and Damaging DNA Effectively

For a DNA-binding fraction of radio-Pt that is more than radio-cisplatin, radio-Pt needs to be modified by DNA-targeting modules. In *chapter 2*, the DNA-intercalator Hoechst33258 labeled with ^{191}Pt and the MYCN gene-targeting PI polyamide (PIP) compound labeled with ^{191}Pt were successfully developed. Interestingly, ^{191}Pt exhibited superior DNA binding and damaging ability when being delivered to DNA using DNA-binding modules. The radio-Pt–based agents [^{191}Pt]Pt-DTPA-Hoechst33258 and [^{191}Pt]Pt-Cys-Hoechst33258 demonstrated more than one order of magnitude greater DNA-binding ability than [^{111}In]In-DTPA-Hoechst3258. [^{191}Pt]Pt-Hoechst33258 labeled via Cys showed a very high DNA-binding fraction and induced extensive DNA damage such as DSBs. Additionally, the novel ^{191}Pt-labeled compound ^{191}Pt-MYCN-PIP targeting the oncogene MYCN bound to DNA efficiently and caused DNA damage, decreasing MYCN gene expression and cell viability, especially in MYCN-amplified Kelly cells. Considering these observations, Auger electron-emitting drugs that target key genes in cell survival may be a novel therapy. Collectively, the two ^{191}Pt-labeled DNA-targeting compounds (Hoechst33258 and MYCN-PIP) successfully increased the DNA-binding fraction of ^{191}Pt, DNA damage like DSBs, and cytotoxicity under certain conditions.

Chapter 3: Development of Radio-Pt Labeled Agents Targeting Tumor Cells In vivo

For *in vivo* uses of radio-Pt-labeled compounds, the two tumor-targeting complexes ^{191}Pt-labeled Nap-Cys-PSMA and Nap-Cys-RGD were developed, and their properties were investigated. ^{191}Pt-labeled Nap-Cys-PSMA and Nap-Cys-RGD kept the specificity of the ligands in the *in vivo* conditions at a certain level. However, the labeled ^{191}Pt to Nap-Cys-PSMA/RGD could remain reactive to other Cys and bound to blood proteins like serum albumin containing Cys residues, which led to a high accumulation of ^{191}Pt in the livers and spleens. ^{191}Pt-labeled agents via multidentate trithiol ligands were intact during blood circulation with low reactivity to protein, suggesting a suitable ligand for ^{191}Pt labeling.

In summary, the present thesis successfully produced n.c.a. ^{191}Pt and ^{191}Pt-labeled compounds, providing insights into the compound design for ^{191}Pt delivery to DNA and tumors. The ^{191}Pt-labeled products expressed the unique DNA-binding ability leading to the high DNA-damaging effect of Auger electrons from ^{191}Pt bound to DNA. These results and findings would contribute to the development of DNA-targeted Auger electron cancer therapy.