Preparation and Evaluation of ^{177}Lu Labeled Nitroimidazole Derivative for Targeting Tumor Hypoxia

Mi-Sun Pyun, Young-Don Hong, Kang-Hyuk Choi, So-Young Lee, Fenelope Felipe, and Sun-Ju Choi
Radioisotope Research and Development Division, Korea Atomic Energy Research Institute
Daedok-daero 1045, Yuseong-gu, Daejon 305-353, Korea
**Corresponding author: mspyun@kaeri.re.kr*

1. Introduction

Hypoxia has been known be an important physiological parameter for tumor growth and has been reported to be the cause for poor response to chemotherapy. Mechanistically, tumor hypoxia results from an imbalance between oxygen delivery and oxygen consumption. The identification and quantification of tumor hypoxia may predict the out come and may identify patients who might benefit from concomitant radiosensitizing therapy to overcome the hypoxia effect.

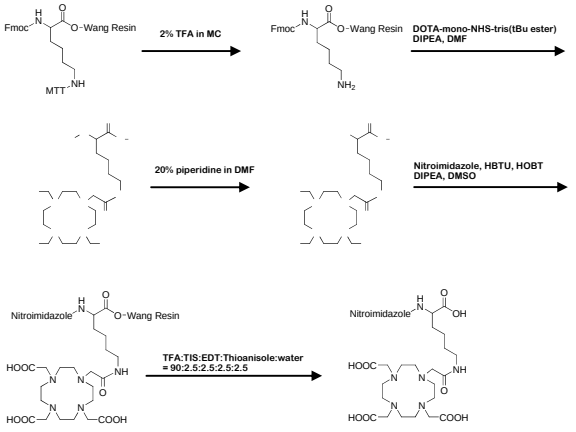
In nuclear medicine, there is an interesting observation that nitroimidazole derivatives can selectively accumulate in the hypoxic regions, a character of a good radiosensitizer.

In this study, we have synthesized a 2-nitroimidazole derivative containing a DOTA chelator by using a solid phase reaction. The nitroimidazole analogue was radiolabeled with ^{177}Lu. Furthermore biodistribution study was performed to evaluate its tissue uptake, *in vivo*.

2. Methods and Results

All chemicals were purchased from Aldrich and used without any further purification. All the processes of the reaction were identified with NMR. $^{177}LuCl_3$ was produced by the KAERI (Korean Atomic Energy Research Institute).

2.1 Synthesis of 2-nitroimidazole-DOTA derivative



Scheme. 1. Reaction pathway of the 2-nitroimidazole-DOTA.

All procedures were done by using solid phase synthesizer. In order to prepare nitroimidazole-DOTA, we first designed and synthesized to introduce DOTA via selective deprotection of the protected lysine-Wang resin. After deprotection of the Fmoc using 20% piperidine, DOTA-Lys-Wang resin was coupled with 2-nitroimidazole and then Wang resin was eliminated under TFA:TIS:EDT:Thioanixole:water condition. Chemical purity was identified with HPLC that showed 2-nitroimidazole-DOTA derivative at 4.4 min.

MS: *m/z* 686$[M+1]^+$

2.2 Radiolabeling

For the radiolabeling, the solution vial was prepared containing 100ug of nitroimidazole-DOTA derivative, 5mg of ascorbic acid and 6mg of dihydroxybenzoic acid dissolved in 1M sodium acetate buffer (pH 5.5). After injecting radioisotope ^{177}Lu (1mCi) to the vial, it was heated for 30min at 90℃ and cooled on ice. HPLC analysis was performed using the gradient system using 0.1% TFA in water(A), 0.1% TFA in ACN(B), Flow rate 1ml/min; 100-90% A in 2min, 90-60% A in 10min, 60-30% A in 2 min, 30-30% A in 3min, 30-100% A in 3 min. The HPLC analysis of the ^{177}Lu-nitroimidazole-DOTA showed a single peak at 6.8min. (Fig.1).

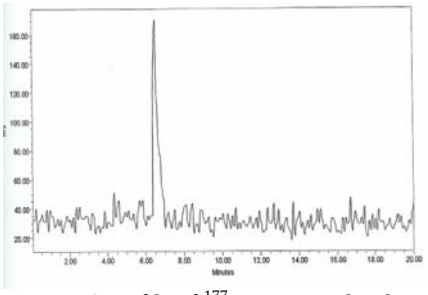


Fig. 1. HPLC profile of ^{177}Lu-nitroimidazole-DOTA.

2.3 Biodistribution study

Biodistribution study was performed on 18-23g female nu/nu Balb/C mice that had been subcutaneously implanted with the 1×10^6 Calu6 human lung cells. Tumors were allowed to grow for 21 d, at which time the animals receive 5uCi of ^{177}Lu labeled nitroimidazole(1ug) in 100ul of saline via lateral tail vein injection. At specified time points (2, 24hr), the tumors, blood and various tissues and organs (heart, liver, spleen, kidney, stomach, small intestine, large intestine, lungs) were removed and weighed, and the

biodistribution was detected using gamma counter. The results show quick renal clearance.

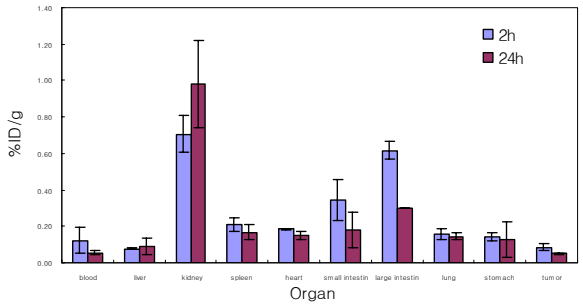


Fig. 2. Biodistribution results at 2 and 24h post-injection of ^{177}Lu-labeled nitromidazole-DOTA.

3. Conclusions

We prepared a new nitroimidazole derivative containing DOTA as a chelating agent of a $^{177}LuCl_3$-precursor for the evaluation of the tumor targeting hypoxia. High radiolabeling efficiency was successfully performed under heating condition. Furthermore, ^{177}Lu-labeled nitromidazole showed quick renal clearance as indicated by the high uptake in the kidneys.

REFERENCES

[1] M. B. Mallia, S. Subramanian, S. Banerjee, H. D. Sarma and M. Venkatesh, Evaluation of $^{99m}Tc(CO)_3$ complex of 2-methyl-5-nitroimidazole as an agent for targeting tumor hypoxia, Bioorganic & Medicinal Chemistry, Vol. 14, p. 7666, 2006.

[2] T. Das, S. Chakraborty, S. Banerjee, A. Mukherjee, G. Samuel, H. D. Sarma, C. K. K. Nair, V. T. Kagiya and M. Venkatesh, Preparation and preliminary biological evaluation of a ^{177}Lu labeled sanazole derivative for possible use in targeting tumor hypoxia, Bioorganic & Medicinal Chemistry, Vol. 12, p. 6077, 2006.

[3] T. Chu, S. Hu, B. Wei, Y. Wang, X. Liu and X. Wanga, Synthesis and biological results of the technetium-99m-labeled 4-nitroimidazole for imaging tumor hypoxia, Bioorganic & Medicinal Chemistry Letters, Vol.14, p.747, 2004.

[4] T. Das, S. Banerjee, G. Samuel, H.D. Sarma, A. Korde, M. Venkatesh, M.R.A. Pillai, ^{99m}Tc-labeling studies of a modified metronidazole and its biodistribution in tumor bearing animal models, Nuclear Medicine and Biology, Vol.30 p.127, 2003.

[5] T. Chu, R. Li, S. Hu, X. Liu, X. Wang, Preparation and biodistribution of technetium-99m-labeled 1-(2-nitroimidazole-1-yl)-propanhydroxyiminoamide (N2IPA) as a tumor hypoxia marker, Nuclear Medicine and Biology, Vol.31 p.199., 2004.

[6] T. Chu, R. Li, S. Hu, X. Liu, X. Wang, Preparation and biodistribution of technetium-99m-labeled 1-(2-nitroimidazole-1-yl)-propanhydroxyiminoamide (N2IPA) as a tumor hypoxia marker, Nuclear Medicine and Biology, Vol.31 p.199, 2004.

[7] B. Gallez, C. Baudelet and Be´ne´dicte F. Jordan, Assessment of tumor oxygenation by electron paramagnetic resonance: principles and applications NMR Biomed., Vol.17, p.240, 2004.