RI内用療法を目指した
シグマ受容体標的化合物の基礎的検討

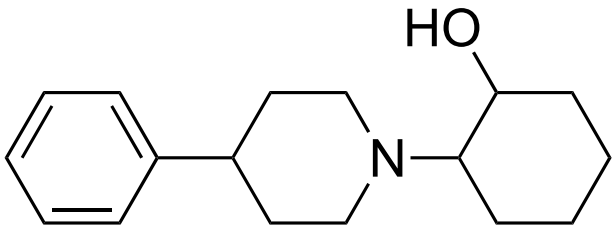
金沢大学医薬保健研究域薬学系
小川　数馬

Introduction

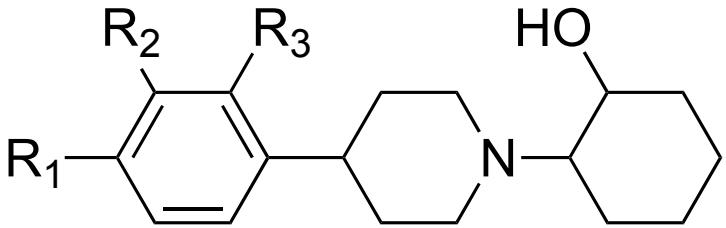
- シグマ受容体は、シグマ-1とシグマ-2といった二つのサブタイプが同定されている(1)
- シグマ受容体は、種々の癌細胞において過剰発現していることが報告されている(2)
- この癌における過剰発現により、シグマ受容体は、癌診断薬や癌治療薬開発のための標的分子と成り得る

(1) Quirion R et al. *Trends Pharmacol Sci* (1992)
(2) Vilner BJ et al. *Cancer Res* (1995)

Chemical Structures of Vesamicol Analogs



Vesamicol



Iodovesamicol (IV)

	R_1	R_2	R_3
o-Iodovesamicol (*o*IV)	I	H	H
m-Iodovesamicol (*m*IV)	H	I	H
p-Iodovesamicol (*p*IV)	H	H	I

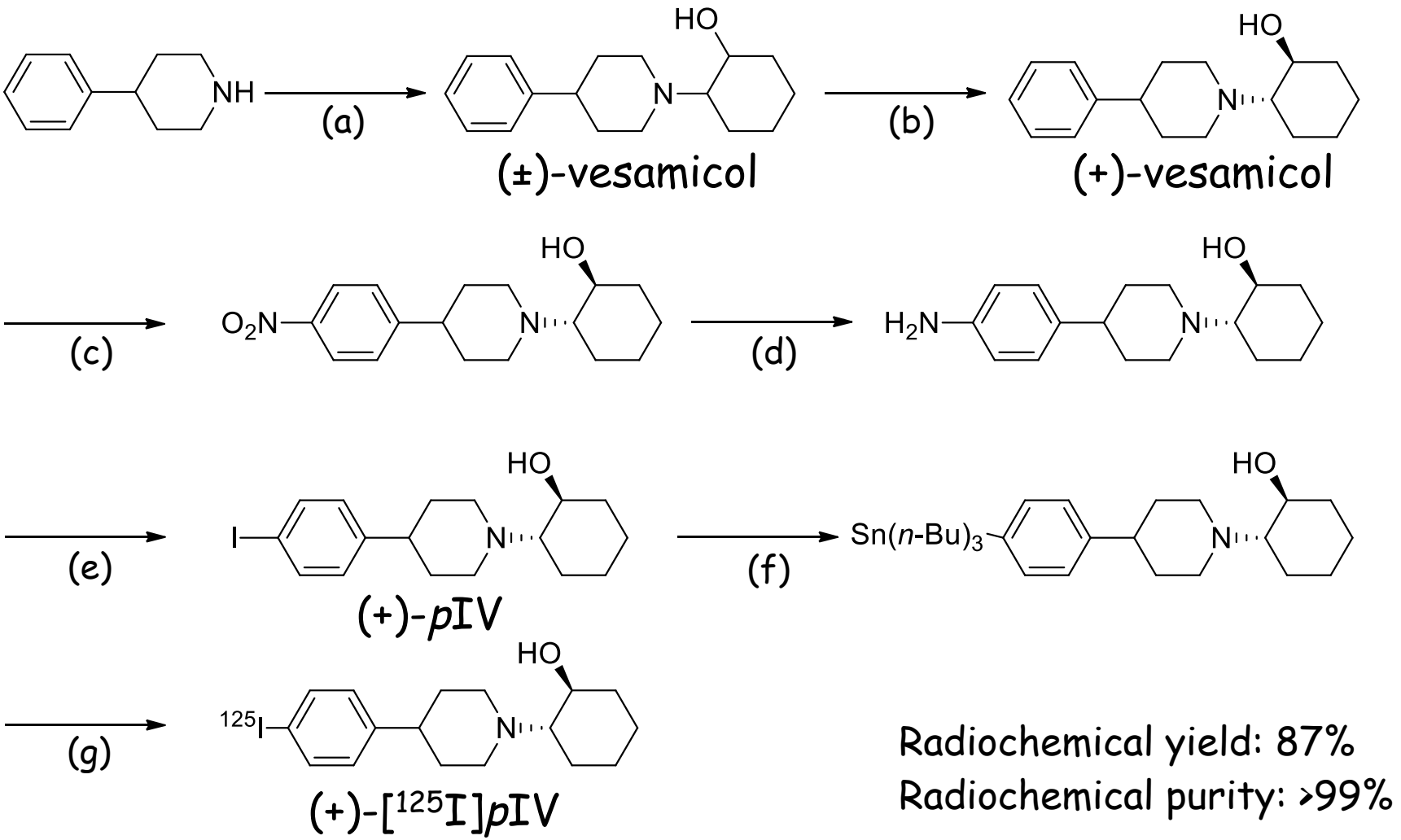
Affinities (nM) of Vesamicol Analogs for Sigma Receptors

	Sigma-1	Sigma-2
(+)-Vesamicol	31.8 ± 11.4	359 ± 39
(-)-Vesamicol	74.9 ± 18.6	421 ± 69
(+)-*o*IV	14.6 ± 1.7	198 ± 93
(-)-*o*IV	62.2 ± 12.0	554 ± 137
(+)-*m*IV	2.5 ± 0.5	40.0 ± 14.7
(-)-*m*IV	4.5 ± 0.5	42.9 ± 15.0
(+)-*p*IV	1.3 ± 0.5	20.4 ± 2.0
(-)-*p*IV	3.4 ± 0.5	28.1 ± 3.9
(+)-Pentazocine	19.9 ± 3.5	2680 ± 162
Haloperidol	13.5 ± 2.0	110 ± 4

*K*i values derived from IC_{50} values accoriding to the equation, $K_i = IC_{50}(1+C/Kd)$. Data are expressed as *K*i (nM) (mean ± SEM).

K Shiba, K. Ogawa et al. *Bioorg Med Chem* (2005)

Preparation of (+)-[^{125}I]*p*IV

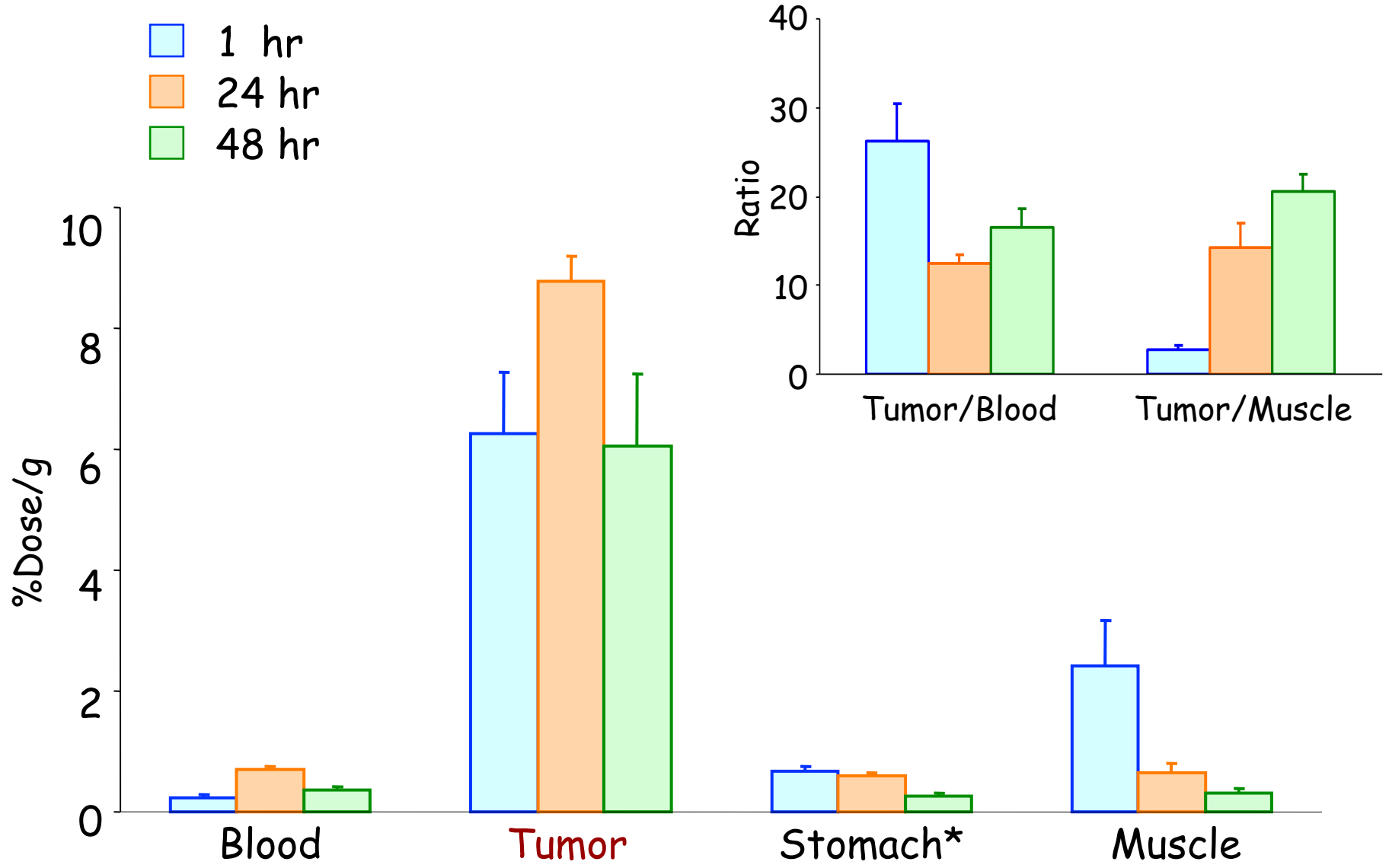


Radiochemical yield: 87%

Radiochemical purity: >99%

(a) cyclohexene oxide. (b) (+)-di-*p*-toluoyl-D-tartaric acid. (c) HNO_3, H_2SO_4.
(d) Fe, HCl. (e) $NaNO_2$, NaI. (f)[$(n\text{-}Bu)_3Sn]_2$, $(Ph_3P)_4Pd$. (g)[^{125}I]NaI, H_2O_2

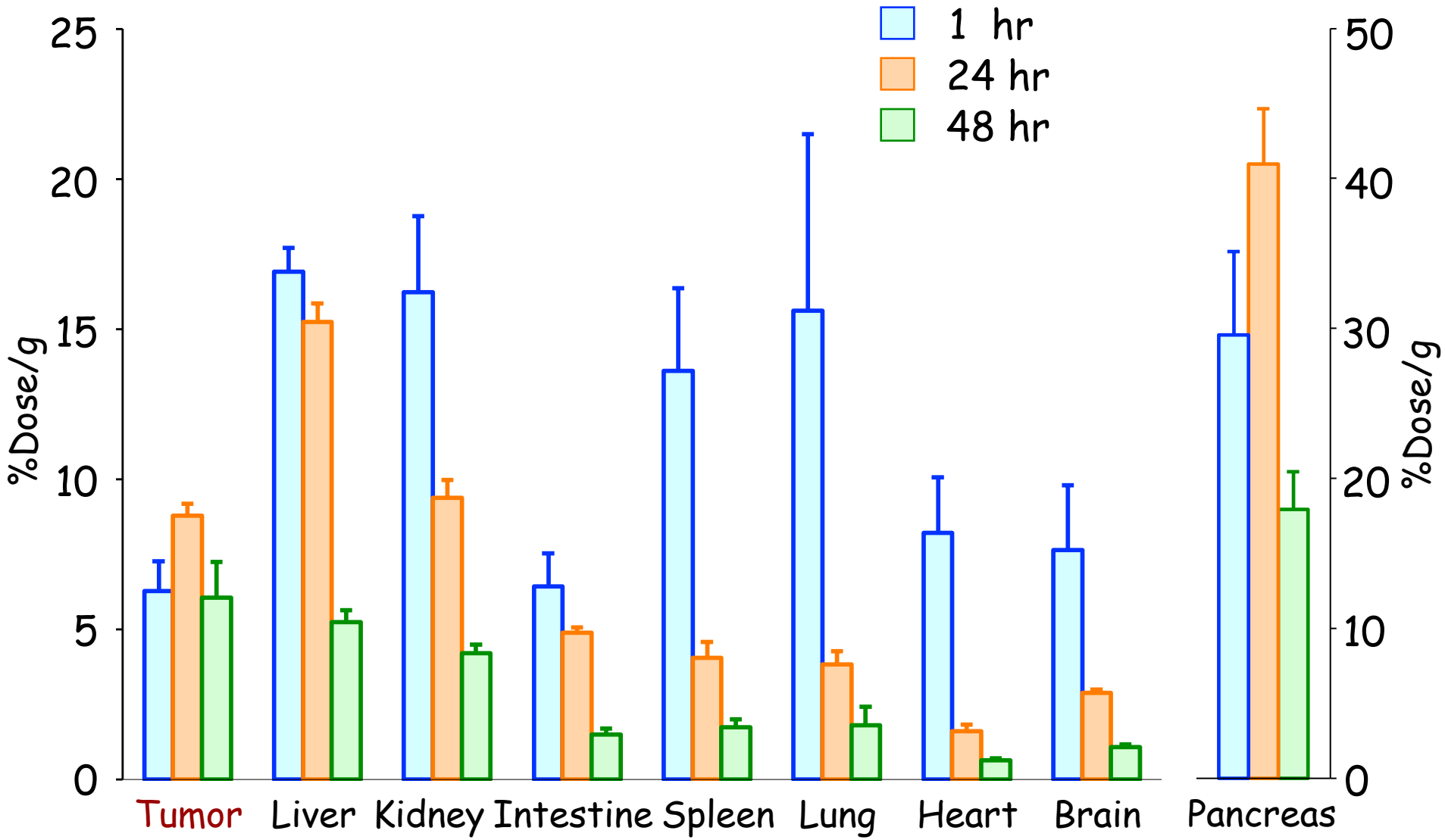
Biodistribution of (+)-[^{125}I]*p*IV in DU-145 Tumor-bearing Mice



*%Dose

K. Ogawa et al. *Cancer Sci*, (2009)

Biodistribution of (+)-[^{125}I]*p*IV in DU-145 Tumor-bearing Mice



K. Ogawa et al. *Cancer Sci*, (2009)

Analysis of Metabolites after Injection of (+)-[^{125}I]*p*IV in Tumor-bearing Mice

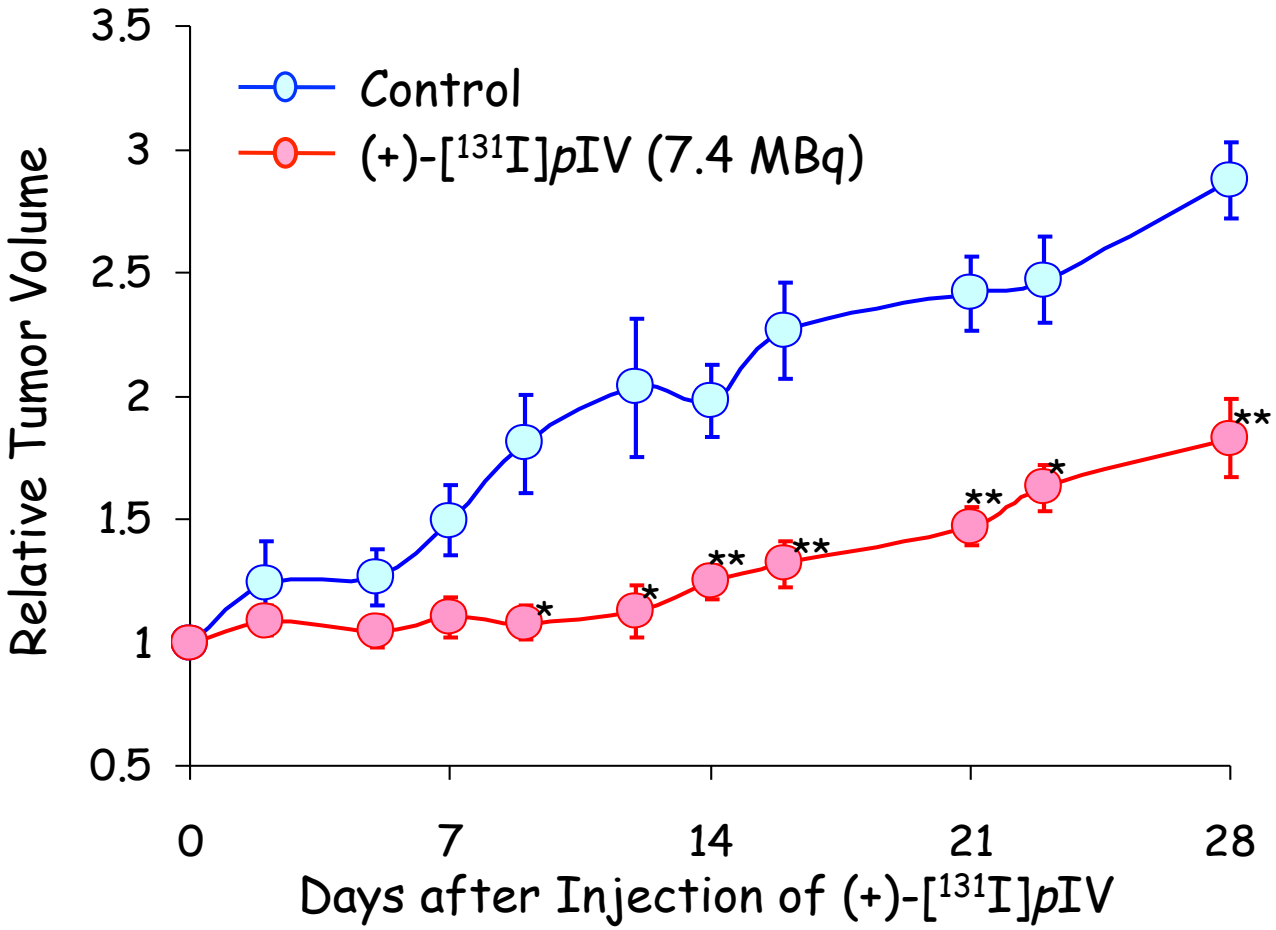
Tissue	Time after Injection	
	1 hour	24 hours
Blood	3.7 (2.6)	1.4 (1.5)
Tumor	83.1 (6.0)	40.7 (2.8)
Liver	76.0 (5.3)	10.4 (2.2)
Kidney	86.5 (5.5)	8.7 (2.0)
Lung	86.2 (4.0)	10.8 (0.8)
Brain	94.8 (2.6)	22.8 (1.3)

Data are expressed as % of intact (+)-[^{125}I]*p*IV.
Each value represents the mean (SD) for three samples.

K. Ogawa et al. *Cancer Sci*, (2009)

Receptor Radionuclide Therapy

腫瘍の大きさをノギスにより測り、腫瘍の体積を Volume = [length x $(width)^2$]/2 により求め、(+)-[^{131}I]*p*IV投与時に対する相対値で評価した

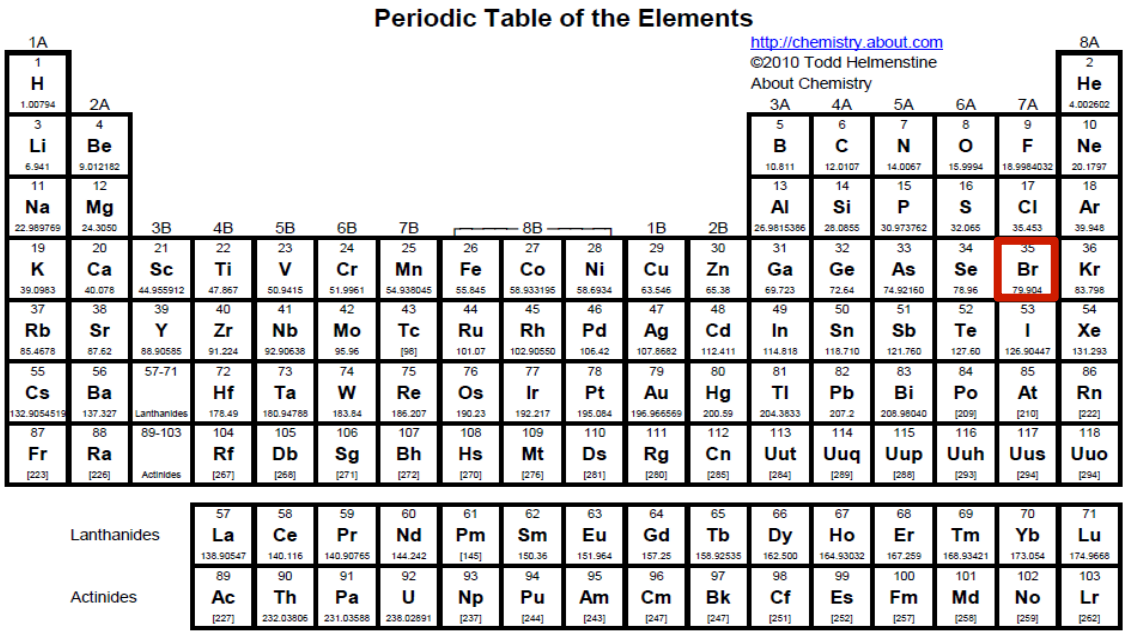


*p < 0.05, **p < 0.01 vs control

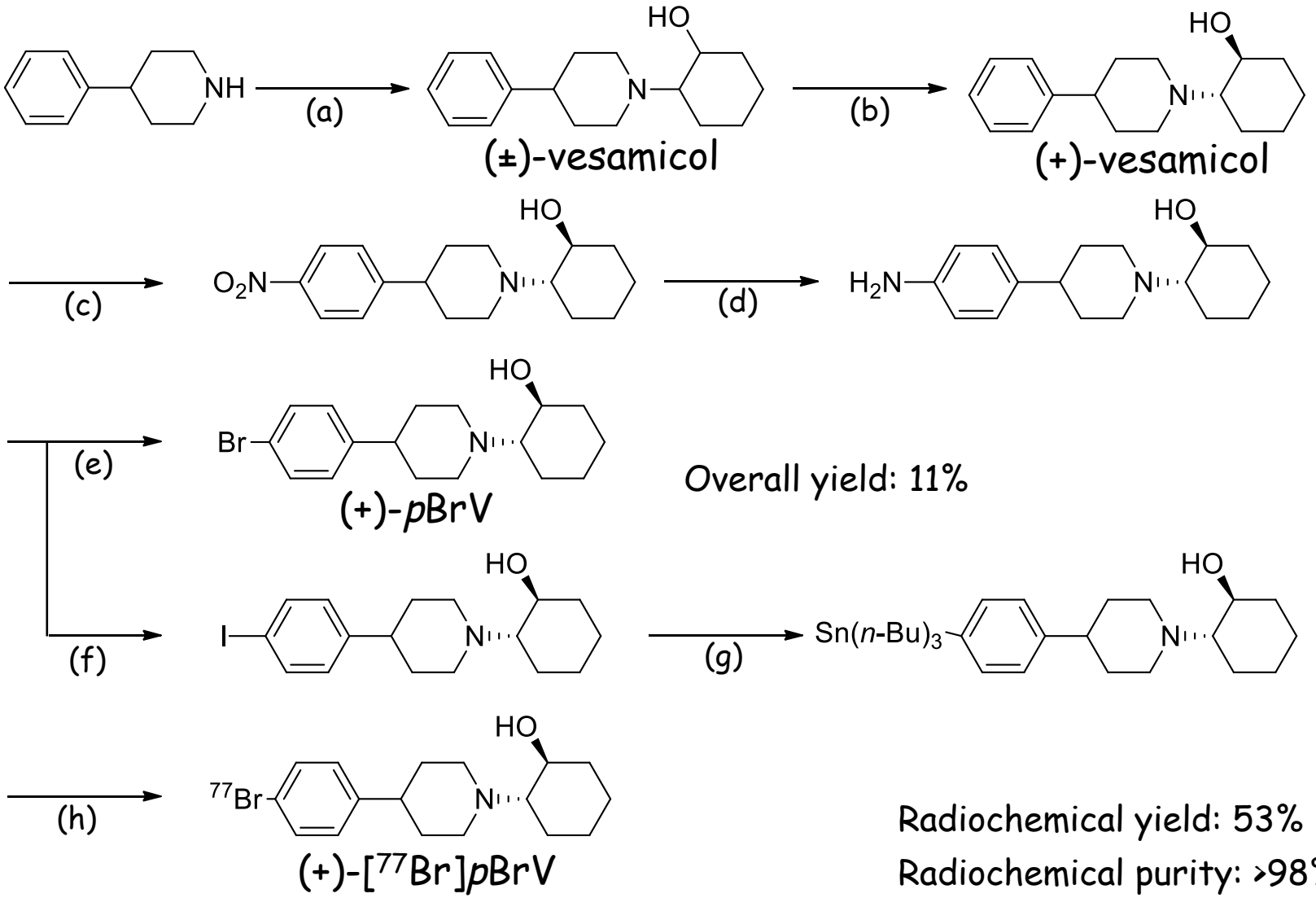
K. Ogawa et al. *Cancer Sci*, (2009)

Bromine Radioisotopes

同位体	^{76}Br	^{77}Br
半減期	16時間	57時間
壊変形式	EC (43%)	EC (99%)
	β^+ (57%)	β^+ (1%)
製造核反応	^{76}Se (p, n) ^{76}Br	^{77}Se (p, n) ^{77}Br



Preparation of (+)-[^{77}Br]*p*BrV



(a) cyclohexene oxide. (b) (+)-di-*p*-toluoyl-D-tartric acid. (c) HNO_3, H_2SO_4. (d) Fe, HCl.
(e) HBr, $NaNO_2$, CuBr (f) $NaNO_2$, NaI. (g)[(*n*-Bu)$_3$Sn]$_2$, $(Ph_3P)_4Pd$. (h)[^{77}I]Br$^-$, chloramine-T

Affinities (nM) of Vesamicol Analogs for Sigma Receptors

	Sigma 1 *K*i (nM)	Sigma 2 *K*i (nM)
(+)-Vesamicol	19.9 ± 2.7	164.7 ± 75.4
(+)-*p*BrV	1.0 ± 0.2	21.2 ± 1.1
(-)-*p*BrV	3.4 ± 0.3	48.9 ± 6.5
(+)-*p*IV	1.6 ± 0.8	14.3 ± 2.4
(+)-Pentazocine	10.2 ± 1.6	2541.8 ± 426.6
Haloperidol	6.1 ± 0.9	56.7 ± 4.8

*K*i values derived from IC_{50} values according to the equation, $K_i = IC_{50}(1+C/Kd)$. Data are expressed as *K*i (nM) (mean ± SEM).

K. Ogawa et al. *Nucl Med Biol*, (2013)

Partition Coefficient

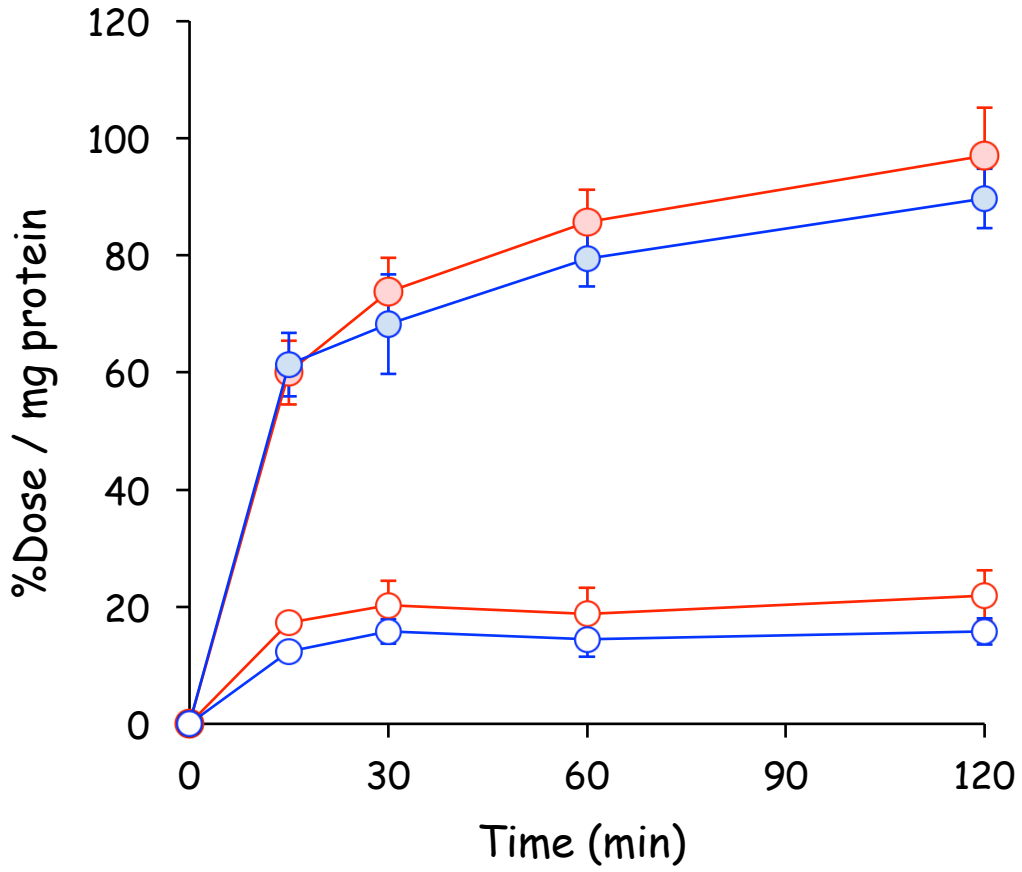
Compound	Log *P*
(+)-[^{125}I]*p*IV	2.08 (0.02)
(+)-[^{77}Br]*p*BrV	1.58 (0.02)

Each value represents the mean (SD) for four samples.

同量の1-オクタノールと0.02 Mリン酸緩衝液(pH 7.4)に(+)-[^{125}I]-IV-OHを加えボルテックス、遠心分離後、1-オクタノールのみをとり、新たに同量の緩衝液を加えた。同様にボルテックス、遠心分離後、有機相、水相の放射能を測定した
分配係数log *P*は log *P* = log (有機相の放射能濃度 / 水相の放射能濃度)で求めた

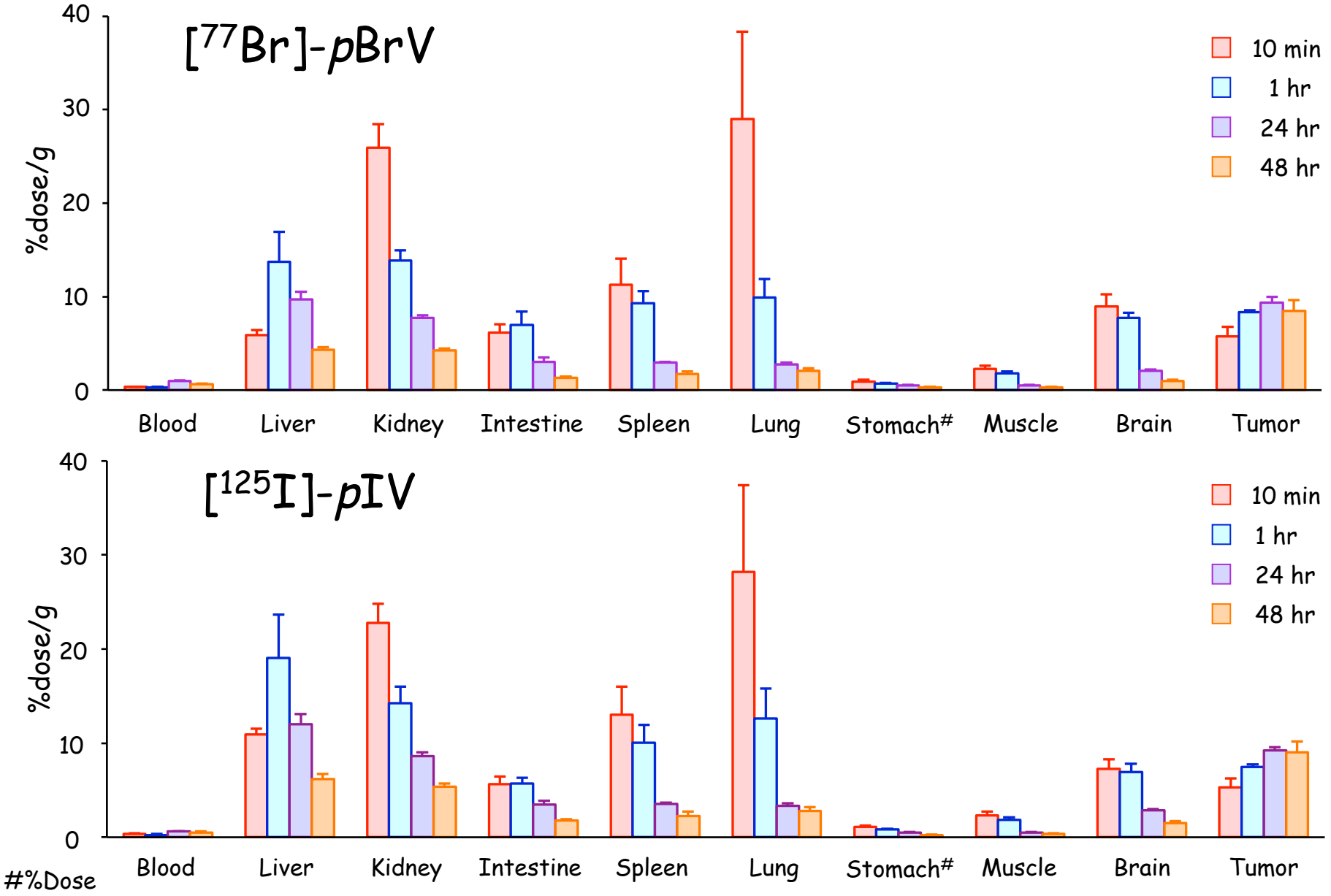
K. Ogawa et al. *Nucl Med Biol*, (2013)

Cell (DU-145) Uptake Study of (+)-[^{77}Br]*p*BrV



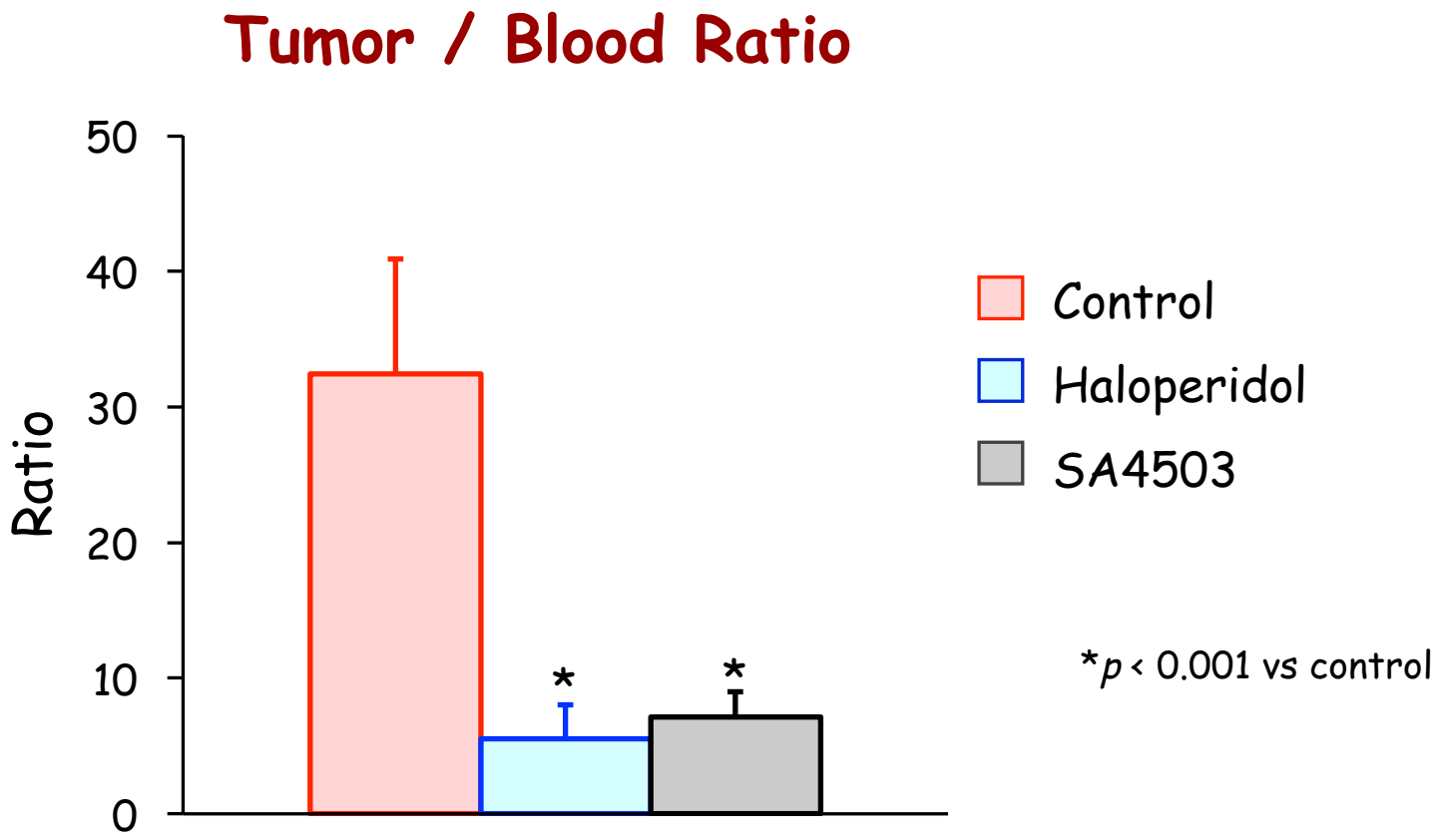
K. Ogawa et al. *Nucl Med Biol*, (2013)

Biodistribution of (+)-[^{77}Br]*p*BrV and (+)-[^{125}I]*p*IV in DU-145 Tumor-bearing Mice



Blocking Study *In Vivo*

DU-145担癌マウスに[^{77}Br]-*p*BrVとシグマリガンド[haloperidol (10 μmol/kg), SA4503 (10 μmol/kg)]を同時静脈内投与した
投与1時間後に屠殺し、腫瘍の重量と放射能を測定した

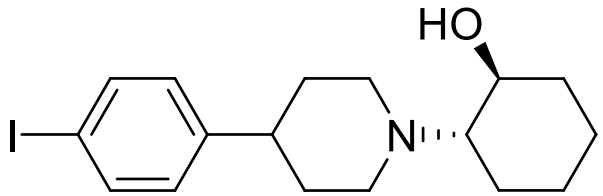


K. Ogawa et al. *Nucl Med Biol*, (2013)

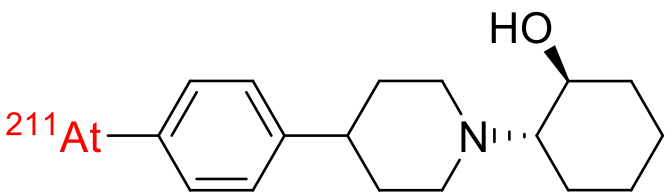
The Aim of This Study

(+)-*p*IVのIを^{211}Atに代替した(+)-[^{211}At]*p*AtVを作製し、
RI内用療法に向けた基礎的検討を行った

- (+)-[^{211}At]*p*AtVの合成
- 分配係数測定
- DU145細胞取込実験
- DU145担癌マウスを用いた体内放射能分布実験

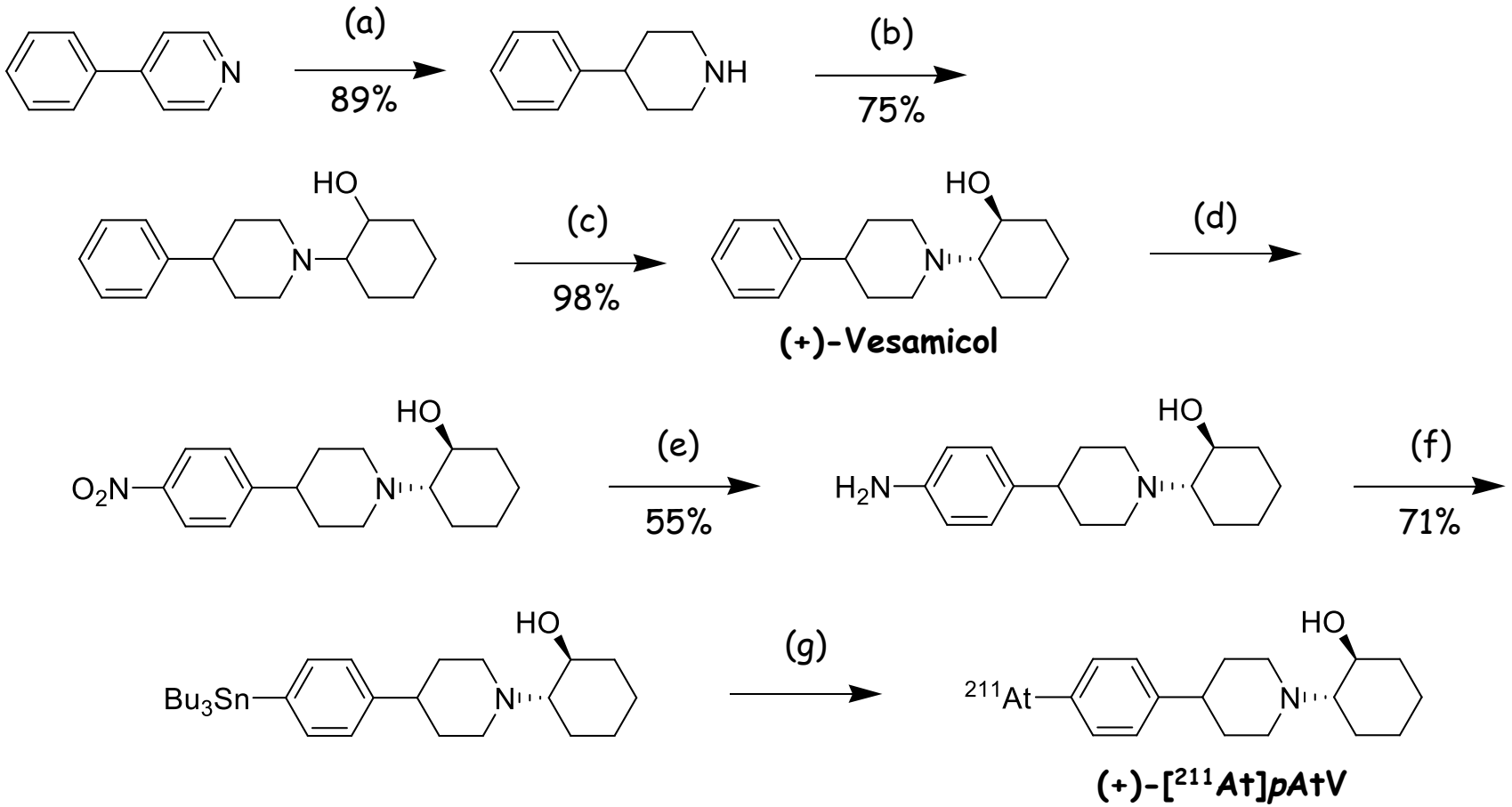


(+)-*p*-Iodovesamicol [(+)-*p*IV]



(+)-*p*-Astatovesamicol [(+)-*p*AtV]

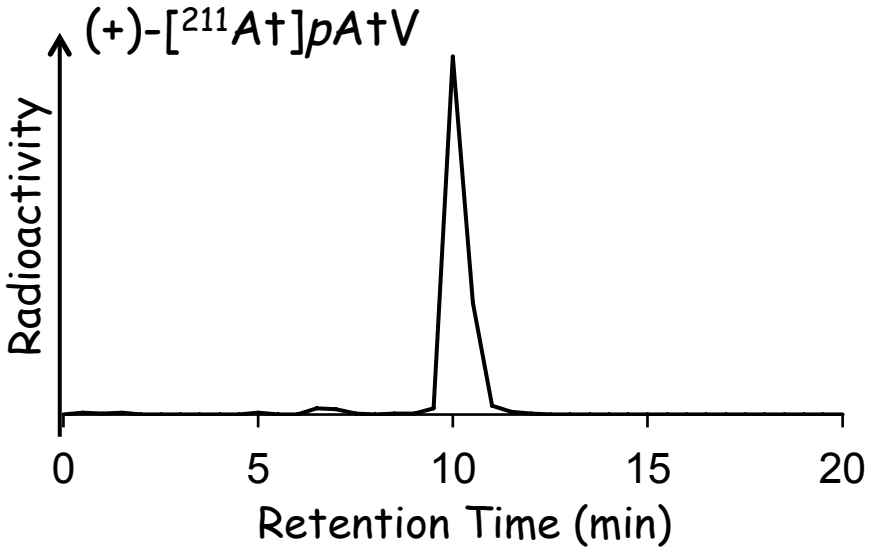
Preparation of (+)-[^{211}At]*p*AtV

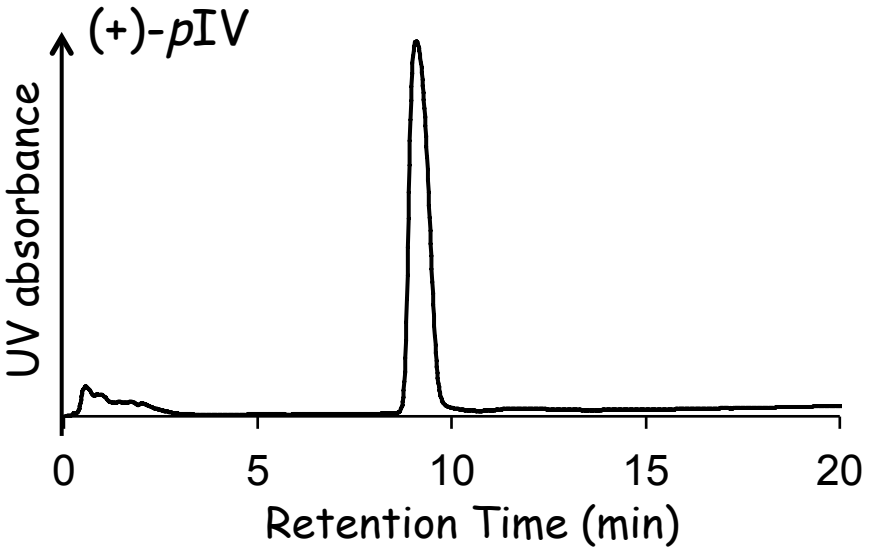


Radiochemical yield : 56%
Radiochemical purity : > 95%

(a) Na; (b) Cyclohexene oxide; (c) (+)-Di-*p*-toluoyl-D-tartaric acid; (d) HNO_3, H_2SO_4; (e) Fe, HCl; (f) I_2, Hexabutylditin; (g) $^{211}At^-$, N-Chlorosccinimide

Chromatogram and Partition Coefficient of (+)-[^{211}At]*p*AtV





HPLC条件
カラム：5C_{18} MS-II 4.6×150 mm
溶出溶媒：CH_3CN:H_2O(0.05% TEAを含む)
75:25→95:5(20 min)

波長：280 nm
流速：1 mL/min

	log *P* value
(+)-[^{211}At]*p*AtV	2.14 ± 0.02
(+)-[^{125}I]*p*IV	2.08 ± 0.11

Each value represents the mean ± SD for four samples.

Summary

- 放射化学的純度95%以上で、(+)-[^{211}At]*p*AtVの標識に成功した
- (+)-[^{211}At]*p*AtVの癌細胞への取込はシグマ受容体特異的なものであった
- (+)-[^{211}At]*p*AtVは投与後早期の腫瘍への高集積および滞留が観察された

(+)-[^{211}At]*p*AtVのRI内用療法への応用の可能性が示された