Pergamon

Bioorganic & Medicinal Chemistry Letters 11 (2001) 1045–1047

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

Synthesis and Structure–Activity Studies of Side-Chain Derivatized Arylhydantoins for Investigation as Androgen Receptor Radioligands

Marcian E. Van Dort* and Yong-Woon Jung

Division of Nuclear Medicine, Department of Radiology, 3480 Kresge III Building, The University of Michigan Medical School, Ann Arbor, MI 48109-0552, USA

Received 22 December 2000; accepted 20 February 2001

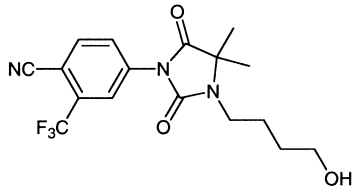
Abstract—A series of arylhydantoin derivatives modeled after the antiandrogen RU 58841 was generated to identify potential candidates for development as androgen receptor (AR) radioligands. Side-chain modified derivatives of RU 58841, suitable for labeling with either carbon-11 or radiohalogens (fluorine-18, iodine-123), were synthesized and tested for their AR binding affinities. The *N*-(iodopropenyl) derivative **13** ($K_i = 13$ nM) is a potential candidate for development as a radioiodinated AR ligand.

The hormone dependency of prostate cancer is well established and androgen receptor (AR) expression is frequently observed in primary prostate tumors and metastases.[1] As a consequence, a variety of radiolabeled AR ligands are under investigation for the noninvasive imaging of tumor sites in prostate cancer using positron emission tomography (PET) or single photon emission computed tomography (SPECT).[2–6] The majority of these studies reported to date have focused on steroid-based radioligands.

The recent emergence of high-affinity, nonsteroidal antiandrogens offers a useful alternative approach towards AR radioligand development.[7–11] As an example, the arylhydantoin derivative RU 58841 (Chart 1) is reported to display high affinity ($K_a = 1.2$ nM) and selectivity for AR (>1000-fold selectivity for AR over progestin, glucocorticoid, mineralocorticoid and estrogen receptors).[10] The excellent AR selectivity, lower lipophilicity and ease of structural modification of RU 58841 and its derivatives as compared to steroid-based ligands make these compounds attractive candidates for investigation as AR radioligands.

In our investigations, RU 58841 displayed nanomolar equilibrium binding affinity ($K_i = 26$ nM) towards the rat AR in radioligand binding studies. Encouraged by this finding, we initiated a study of RU 58841 derivatives to identify high-affinity analogues for subsequent radiotracer development. Our radiosynthetic strategy envisaged introduction of the radiolabel at the hydantoin *N*(3)-position via attachment to an appropriate spacer group. Accordingly, a series of side-chain modified derivatives of RU 58841 (Table 1) suitable for labeling with either PET (carbon-11, fluorine-18) or SPECT (iodine-123) radioisotopes was synthesized for evaluation of their in vitro AR binding affinities.

RU 58841 and its key intermediate 4-(4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile (**1**) were synthesized as previously reported.[10] Synthesis of compounds **2**–**8** was conducted as shown in Scheme 1 via generation of the anion of **1** by treatment with NaH in DMF and subsequent alkylation with the appropriate commercially available alkyl bromide or iodide.[12]



RU 58841

Chart 1.

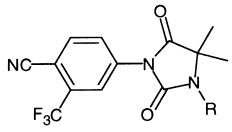
*Corresponding author. Tel.: +1-734-763-4647; fax: +1-734-936-1571; e-mail: mvandort@umich.edu

0960-894X/01/$ - see front matter
PII: S0960-894X(01)00146-9

Compounds **9**–**12** were synthesized in a similar fashion from the appropriate iodophenylalkyl tosylate precursors.[13] The *E*-isomer of the iodopropenyl derivative (**13**) was synthesized by alkylation of **1** with the *E*-isomer of 1-(tri-*n*-butylstannyl)-3-chloro-1-propene[14] and treatment of the resulting intermediate (**13a**) with iodine in chloroform (Scheme 2). The pure products were isolated in 50–95% yields after chromatography and recrystallized from EtOAc/hexane mixtures. The compounds gave ^{1}H NMR and elemental analysis or mass spectrometry data consistent with the assigned structures.

The binding affinities of new ligands and reference compounds to the rat prostate cytosolic AR were determined using a competitive binding assay in the presence of the high-affinity AR radioligand, [^{3}H]mibolerone.[15–17] These data (expressed as inhibition constants, K_i) are presented in Table 1. All ligands demonstrated monophasic radioligand displacement curves (Hill coefficient close to unity) indicating interaction with a single class of binding sites. The lead derivative RU 58841 displayed a K_i value of 26 nM in these assays.

Table 1. Inhibition constants (K_i) for ligands at the rat androgen receptor

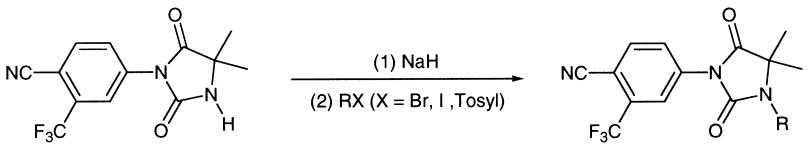


Compound	R	K_i (nM)[a]	mp (°C)[b]	Yield (%)[c]
2	CH_3	37±5	153–154	95
3	$(CH_2)_2OCH_3$	120±20	Oil	89
4	$(CH_2)_2F$	141±31	105–106	59
5	$(CH_2)_3F$	321±40	91–93	90
6	$(CH_2)_4F$	213±27	71–73	86
7	CH_2(*m*-I-Ph)	123±25	113–114	91
8	CH_2(*p*-I-Ph)	150±10	101–103	86
9	$(CH_2)_2$(*m*-I-Ph)	809±120	146–148	50
10	$(CH_2)_2$(*p*-I-Ph)	415±82	144–145	89
11	$(CH_2)_3$(*m*-I-Ph)	360±20	90–92	56
12	$(CH_2)_3$(*p*-I-Ph)	65±15	112–113	78
13	(*E*)-CH_2CH=CHI	13±2	108–109	91
RU 58841	$(CH_2)_4OH$	26±5		
Testosterone		4.9±1.8		
Mibolerone		0.75±0.08		

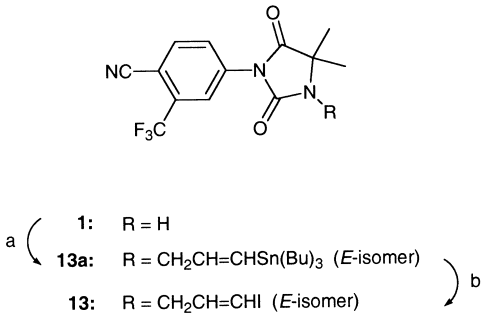
[a]Data are presented as mean±SEM of three independent determinations each conducted in duplicate.
[b]Solids gave white crystals after recrystallization from EtOAc/hexane (1:5).
[c]Isolated yields after chromatography.

The *N*-methyl and *N*-(2-methoxyethyl) substituted hydantoin derivatives (**2** and **3**) were synthesized as potential leads for the development of carbon-11 labeled AR radioligands. The AR binding affinity of the *N*-methyl analogue **2** (K_i = 37 nM) was comparable to that of the lead compound RU 58841 (K_i = 26 nM) suggesting that the small-volume methyl group was well tolerated at the *N*(3) position. However, the 2-methoxyethyl substituent was not as well tolerated displaying an almost 5-fold reduction in AR affinity. A series of RU 58841 derivatives bearing fluoroalkyl side chains (**4**–**6**) was also prepared to identify candidates for labeling with fluorine-18. Since fluorine can mimic a hydroxyl group in some biologically-active compounds,[18] we were particularly interested in the effect of this substitution in RU 58841. All of the fluoroalkyl derivatives displayed weaker AR binding affinity than RU 58841. Notably, replacement of the hydroxyl group of RU 58841 with fluorine (**6**) leads to an 8-fold reduction in its AR binding affinity. Since fluorine can only function as a hydrogen bond acceptor,[19] the enhanced binding affinity of RU 58841 over **6** may be due to a hydrogen bond donor interaction of its hydroxyl group with the receptor site. However, it is also possible that the lower binding affinity of **6** may be due to an unfavorable hydrophobic interaction of fluorine with the AR binding site.

In our design of iodinated compounds, we focused solely on derivatives having an sp^2 carbon-bound iodine functionality as these are shown to have a higher metabolic stability than iodoalkyl derivatives.[20,21] Accordingly, a series of *N*-(iodophenylalkyl) derivatives (**7**–**12**) and the *N*-(iodopropenyl) derivative **13** (*E*-isomer) were synthesized for AR binding studies. A broad spread of AR affinities was observed within the *N*-(iodophenylalkyl) series. Thus, introduction of either a *meta*-iodobenzyl or *para*-iodobenzyl group at *N*(3) (**7** and **8**) led to a 5- to 6-fold reduction in AR affinity, whereas increasing the chain length by a single methylene unit (**9** and **10**) resulted in a dramatic (16- to 32-fold) loss in binding affinity. Interestingly, in the iodophenylpropyl series, the *para*-iodo substituted analogue **12** (K_i = 65 nM) showed a 6-fold improvement in AR affinity over the corresponding *meta*-iodo substituted analogue (**11**). Among the new compounds studied, the *N*-(iodopropenyl) analogue **13** displayed the highest AR binding affinity (K_i = 13 nM), which is a 2-fold improvement over the lead derivative RU 58841. High AR binding affinity (relative binding affinity = 46% of testosterone), has also been reported for the *N*-(cyanomethyl) derivative RU 58642 (R = CH_2CN).[22] The enhanced binding affinity displayed by RU 58642 and **13** may be due to a



Scheme 1.



Scheme 2. (a) (i) NaH/DMF; (ii) (*E*)-$ClCH_2CH{=}CHSn(Bu)_3$; (b) I_2/$CHCl_3$.

favorable π electronic interaction of the cyano and olefinic moieties, respectively, with the AR binding site. Additional studies are planned to confirm this hypothesis.

In summary, our studies suggest that the nature of the side chain in RU 58841 derivatives plays a major role in its AR affinity. In general, introduction of fluoroalkyl or iodophenylalkyl substituents at the *N*(3) position of RU 58841 has a detrimental effect on AR affinity. Such compounds are therefore unsuitable for development as AR radioligands. The *N*-(iodopropenyl) derivative **13**, however, emerges as a new lead for the development of high-affinity radioiodinated AR radioligands. Studies are underway to synthesize [I-123]-labeled **13** for evaluation as a SPECT AR radioligand.

Acknowledgements

We thank the staff of the Phoenix Memorial Laboratories for use of their facilities. This research was supported by grants from the National Institutes of Health (CA 77287) and the SPORE in Prostate Cancer (P50 CA 69568).

References and Notes

1. Hobisch, A.; Culig, Z.; Radmayr, C.; Bartsch, G.; Klocker, H.; Hittmair, A. *Prostate* **1996**, *28*, 129.
2. Katzenellenbogen, J. A. *J. Nucl. Med.* **1995**, *36*, 8S.
3. Welch, M. J.; Downer, J. B.; Katzenellenbogen, J. A. In *Current Directions in Radiopharmaceutical Research and Development*; Mather, S. J., Ed.; Kluwer: Dordrecht, 1996; pp 137–156.
4. Ali, H.; Rousseau, J.; van Lier, J. E. *J. Steroid Biochem. Mol. Biol.* **1994**, *49*, 15.
5. Labaree, D. C.; Brown, T. J.; Hoyte, R. M.; Hochberg, R. B. *J. Nucl. Med.* **1997**, *38*, 402.
6. Cutler, P. D.; Dehdashti, F.; Siegel, B. A.; Downer, J. B.; Welch, M. J. *J. Nucl. Med.* **1996**, *37*, 87P.
7. Dalton, J. T.; Mukherjee, A.; Zhu, Z.; Kirkovsky, L.; Miller, D. D. *Biochem. Biophys. Res. Commun.* **1998**, *244*, 1.
8. Hamann, L. G.; Mani, N. S.; Davis, R. L.; Wang, X.-N.; Marschke, K. B.; Jones, T. K. *J. Med. Chem.* **1999**, *42*, 210.
9. Zhi, L.; Tegley, C. M.; Marschke, K. B.; Jones, T. K. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1009.
10. Battmann, T.; Bonfils, A.; Branche, C.; Humbert, J.; Goubet, F.; Teutsch, G.; Philibert, D. *J. Steroid Biochem. Mol. Biol.* **1994**, *48*, 55.
11. Teutsch, G.; Goubet, F.; Battmann, T.; Bonfils, A.; Bouchoux, F.; Cerede, E.; Gofflo, D.; Gaillard-Kelly, M.; Philibert, D. *J. Steroid Biochem. Mol. Biol.* **1994**, *48*, 111.
12. The preparation of **8** is illustrative. Dry NaH (12 mg, 0.4 mmol) was added to a solution of **1** (99 mg, 0.33 mmol) in dry DMF (1 mL) under argon. The mixture was stirred at ambient temperature for 15 min and treated dropwise with a solution of 4-iodobenzyl bromide (117 mg, 0.4 mmol) in dry DMF (1 mL). Stirring was continued at ambient temperature for 3 h and the reaction monitored by TLC analysis (silica; 35% EtOAc in hexane). The mixture was poured into saturated brine (25 mL) and extracted with ether (2×25 mL). The combined ether extracts were washed successively with saturated brine (25 mL), water (2×25 mL) and dried (Na_2SO_4). The residue obtained after removal of solvent was purified by flash chromatography (35% EtOAc in hexane) to afford 146 mg (86%) of **8** as a white solid: mp 101–103 °C (EtOAc/hexane (1:5)); ^1H NMR ($CDCl_3$) δ 8.17 (s, 1H), 8.03 (d, 1H, $J=8.4$ Hz), 7.93 (d, 1H, $J=8.4$ Hz), 7.69 (d, 2H, $J=8.3$ Hz), 7.13 (d, 2H, $J=8.3$ Hz), 4.56 (s, 2H), 1.43 (s, 6H).
13. Van Dort, M. E.; Kilbourn, M. R.; Chakraborty, P. K.; Richfield, E. K.; Gildersleeve, D. L.; Wieland, D. M. *Appl. Radiat. Isotop.* **1992**, *43*, 671.
14. Elmaleh, D. R.; Fischman, A. J.; Shoup, T. M.; Byon, C.; Hanson, R. N.; Liang, A. Y.; Meltzer, P. C.; Madras, B. K. *J. Nucl. Med.* **1996**, *37*, 1197.
15. Androgen receptor binding assays were conducted by a commercial laboratory (MDS Panlabs, Bothell, WA, USA).
16. Schilling, K.; Liao, S. *Prostate* **1984**, *5*, 581.
17. Liao, S.; Witte, D.; Schilling, K.; Chang, C. *J. Steroid Biochem.* **1984**, *20*, 11.
18. Card, P. J. *J. Carbohydr. Chem.* **1985**, *4*, 451.
19. Brettle, R.; Cross, R.; Frederickson, M.; Haslam, E.; MacBeath, F. S.; Davies, G. M. *Bioorg. Med. Chem. Lett.* **1996**, *6*, 1275.
20. Seevers, R. H.; Counsell, R. E. *Chem. Rev.* **1982**, *82*, 575.
21. Musachio, J. L.; Lever, J. R. *Bioconjugate Chem.* **1992**, *3*, 167.
22. Battmann, T.; Branche, C.; Bouchoux, F.; Cerede, E.; Philibert, D.; Goubet, F.; Teutsch, G.; Gaillard-Kelly, M. *J. Steroid Biochem. Mol. Biol.* **1998**, *64*, 103.