Pergamon

Bioorganic & Medicinal Chemistry Letters 12 (2002) 2607–2609

BIOORGANIC &
MEDICINAL
CHEMISTRY
LETTERS

Synthesis and Characterization of a New RXR Agonist Based on the 6-*tert*-Butyl-1,1-dimethylindanyl Structure

Beatriz Domínguez,[a] M. Jesús Vega,[a] Fredy Sussman[b] and Angel R. de Lera[a,*]

[a]*Departamento de Química Orgánica, Facultad de Ciencias, Universidade de Vigo, 36200 Vigo, Spain*
[b]*Departamento de Química Orgánica, Facultad de Química, Universidade de Santiago de Compostela,*
15706 Santiago de Compostela, Spain

Received 20 March 2002; revised 7 June 2002; accepted 17 June 2002

Abstract—A new ligand for RXR is described, which is based on a 6-*tert*-butyl-1,1-dimethylindanyl skeleton as bioisostere of the hydrophobic retinoid region. The Stille cross-coupling reaction allowed the attachment of the polyene side chain to the indanyl ring. Docking studies were carried out to explain the RXR binding profile of this analogue. © 2002 Elsevier Science Ltd. All rights reserved.

Two families of retinoid receptors,[1,2] which belong to the nuclear receptor superfamily,[3] have been characterized, RARs (isotypes α, β, γ)[1] and RXRs (isotypes α, β, γ).[2] Whereas RAR is activated by both retinoic acid **1** and 9-*cis*-retinoic acid **2**, RXR binds selectively the latter.[4] The regulation of gene transcription by the superfamily of nuclear receptors is a complex sequence of events, requiring activation of specific hormone response elements (HREs) on DNA. A group of nuclear receptors (among them RAR, VDR, PPAR, TR, LXR) share a similar mechanism of transcriptional regulation since they activate HREs as heterodimers having RXR as common partner.[4] This finding has fueled a great interest on the discovery of RXR-selective ligands. In this regard, much effort has been devoted to the synthesis and evaluation of analogues with increased stability by incorporating some of the polyene double bonds present in the side chain of the sensitive native retinoids (**1** and **2**) into aromatic rings.[5] Most commonly the β-ionone ring and the adjacent double bond are replaced by a lipophilic bioisostere, such as a 5,6,7,8-tetrahydro-5,5,8,8-tetramethylnaphthalene (compounds **3**,[6] and **5**,[7] see Fig. 1) and a 1,3-di-*tert*-butylbenzene skeleton (**4**[8]). Structure–activity relationship studies of the 5,6,7,8-tetrahydro-5,5,8,8-tetramethylnaphthalene-based retinoids has revealed the role that steric interactions at the naphthalene 3-position have in receptor selectivity and potency. In this regard LGD1069 **6**,[9] a RXR-selective activator exhibiting greater potency than its 3-demethyl analogue **5**,[7] has recently been approved (bexarotene, Targretin®) for the treatment of cutaneous T-cell lymphoma and AIDS-related Kaposi sarcoma.

Herein, we describe the synthesis, molecular modeling and RXR binding activity of a new retinoid (**7**), featuring a 6-*tert*-butyl-1,1-dimethylindanyl moiety connected to a trienyl carboxylic acid. The hydrophobic unit might be considered structurally similar to a 1,3-di-*tert*-butylphenyl group (see, **4**) but incorporating a methyl group *ortho* to the side chain (as present in **6**).

Retinoid **7** was synthesized as shown in Scheme 1. The key step on the synthesis features a palladium-catalyzed cross-coupling of dienylorganotin derivative **8**[10] and vinyliodide **11** (Stille reaction).[11] The highly stereoselective nature of this coupling ensures in principle retention of the double-bond geometries of both coupling partners. Internal iodide **11** was prepared from alkyne **10**, itself derived from commercially available methylketone **9** through application of Negishi's one-pot procedure,[12] by hydroiodination using the 'in situ' NaI/Me_3SiCl method.[13] The coupling step was challenging due to the low reactivity of internal iodides with stannanes.[11] Even exploiting the rate-acceleration effect enforced by the combination of 'ligandless' palladium and a poorly coordinating ligand (Farina's conditions, Pd_2dba_3, $AsPh_3$, NMP)[14] the coupling required heating to 60 °C to provide access to methylester **12** in 83%

---

*Corresponding author. Tel.: +34-986-812316; fax: +34-986-812556; e-mail: qolera@uvigo.es

0960-894X/02/$ - see front matter
PII: S0960-894X(02)00477-8

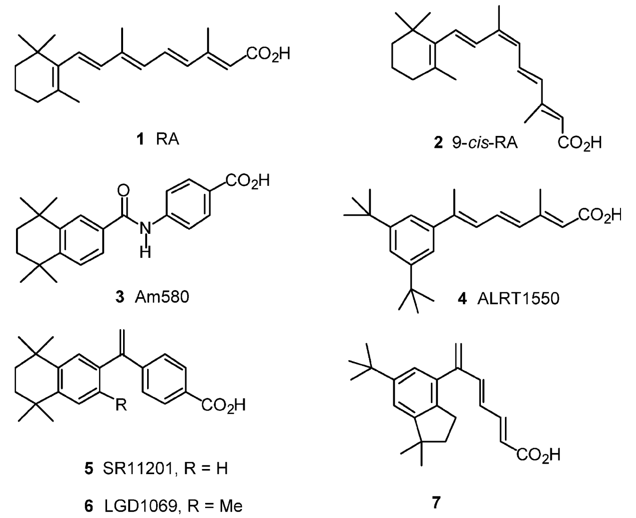
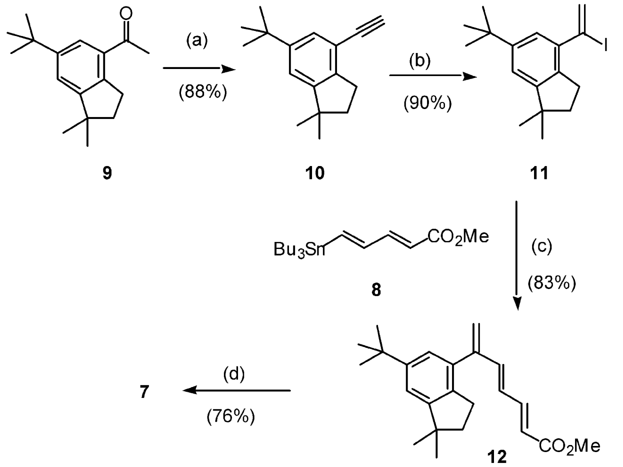


Figure 1. Selected native and synthetic retinoid-receptor ligands.



Scheme 1. Reagents and conditions: (a) (1) LDA, THF, −78 °C; (2) $Cl(O)P(OEt)_2$; (3) LDA, THF, −78 to 25 °C; (b) NaI, Me_3SiCl, CH_3CN; (c) $Pd_2(dba)_3$, $AsPh_3$, NMP; (d) 5 M KOH, MeOH, 90 °C, 10 min.

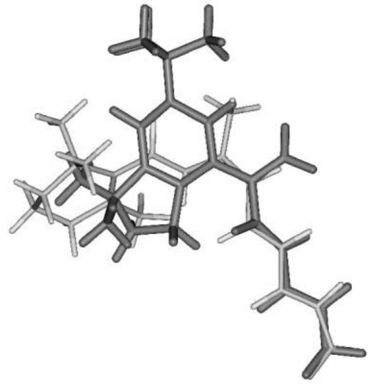


Figure 2. Structure of the indanyl ligand **7** (dark lines) in the close to *s-cis* conformation, superimposed on that of 9-*cis*-retinoic acid **2**.

yield. Saponification then provided the desired carboxylic acid **7** (Scheme 1).

Compound **7** was screened in transactivation assay with co-transfected RXRα, which measures the ability of compounds to activate gene expression at the receptor and reflects the compound's functional activity.[15] It showed an AC_{50} value of 79 (for comparison, the value for LGD1069 **6** is 28). In addition, the competitive binding assay characterizes the ligand's ability to bind to the receptor. The RXRα binding assay was performed as previously described.[16] The dissociation constant (K_d value) was determined as 220 nM (9-*cis*-retinoic acid **2**: $K_d = 50$ nM; LGD1069 **6**: $K_d = 36$ nM).

The molecular structure of the 6-*tert*-butyl-1,1-dimethylindanyl analogue **7** was built using the Builder module of the InsightII suite of programs.[17] Recently, the structure of the RXRα receptor complexed with its natural ligand, 9-*cis*-retinoic acid **2** has been determined by X-ray crystallography.[18] To obtain a model for the binding of ligand **7** to RXRα, we have docked a set of 100 conformers obtained by MD to the receptor bound to its natural ligand, 9-*cis*-retinoic acid **2**, by superimposing the unsaturated chains of both ligands. A set of complexes between the receptor and the indanyl derivative **7** was obtained by removing 9-*cis*-retinoic acid **2**. The resulting structures were energy minimized using 1000 steps of steepest descent method (SD) with the CHARMM force field,[19] a process performed in two stages. In the first part a 100 step SD minimization was run with the ligand frozen, followed by 900 steps of SD minimization for the whole complex. The affinity ranking of the different conformers was evaluated by their interaction energy with RXRα, using the CHARMM force field. To determine the actual conformational variability of the ligand, we run a 0.1 ns simulation of the ligand in vacuum, at 300 K with no constraints, generating a set of 100 conformers, equally spaced in time. The biggest conformational variability (observed in the MD trajectory of this ligand) is the one present in the torsional angle between the indanyl aromatic ring and the triene side chain, which we found to exhibit large departures from planarity. The shallow dependence of the energy surface (on the ring side-chain torsional angle) has been observed before for model molecules that contain a benzene ring with side chain vinyl groups like styrene and stilbene.[20] The computed variability of this dihedral angle confirms the NOE correlations measured by NMR in solution, which indicate the existence of a mixture of *s-cis* and *s-trans* conformers about this bond. One of the extreme conformations is shown in Figure 2, superimposed to the structure of 9-*cis*-retinoic acid **2**.

Based on the Molecular Modeling studies, work is in progress to design and synthesize derivatives of **7** with optimized receptor binding properties.

Acknowledgements

We thank DGICYT (Grant SAF98–0134), *Xunta de Galicia* (Grant PGIDT99PXI30105B, Visiting Research Contract to F. Sussman) and Galderma R&D for financial support. We wish to thank Drs. Uwe Reichert, Philippe Diaz and Serge Michel (Galderma R&D) for the RXRα-binding studies.

References and Notes

1. Petkovich, M.; Brand, N. J.; Kurst, A.; Chambon, P. *Nature* **1987**, *330*, 444.
2. Heyman, R. A.; Mangelsdorf, D. A.; Dyck, J. A.; Stein, R. B.; Eichele, G.; Evans, R. M.; Thaller, C. *Cell* **1992**, *68*, 397.
3. For reviews, see: (a) Evans, R. M. *Science* **1988**, *240*, 889. (b) Mangelsdorf, D. A.; Evans, R. M. *Cell* **1995**, *83*, 841. (c) Rosen, J.; Day, A.; Jones, T. K.; Jones, E. T. T.; Nadzan, A. M.; Stein, R. B. *J. Med. Chem.* **1995**, *38*, 4855. (d) Katzenellenbogen, J. A.; Katzenellenbogen, B. S. *Chem. Biol.* **1996**, *3*, 529. (e) Weatherman, R. V.; Fletterick, R. J.; Scanlan, T. S. *Annu. Rev. Biochem.* **1999**, *68*, 559. (f) Bourquet, W.; Germain, P.; Gronemeyer, H. *Trends Pharmacol. Sci.* **2000**, *21*, 381.
4. Levin, A. A.; Sturzenbecker, L. J.; Kazmer, S.; Bosakowski, T.; Huselton, C.; Allenby, G.; Speck, J.; Kratzeisen, Cl.; Rosenberger, M.; Lovey, A.; Grippo, J. F. *Nature* **1992**, *355*, 359.
5. For recent selected examples, see: (a) Vuligonda, V.; Lin, Y.; Thacher, S. M.; Standeven, A. M.; Kochar, D. M.; Chandraratna, R. A. S. *Bioorg. Med. Chem.* **1999**, *7*, 263. (b) Vuligonda, V.; Chandraratna, R. A. S. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 2289. (c) Vuligonda, V.; Standeven, A. M.; Escobar, M.; Chandraratna, R. A. S. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 743. (d) Canan Koch, S. S.; Dardashti, L. J.; Cesario, R. M.; Croston, G. E.; Boehm, M. F.; Heyman, R. A.; Nadzan, A. M. *J. Med. Chem.* **1999**, *42*, 742. (e) Dawson, M. I.; Hobbs, P. D.; Jong, L.; Xiao, D.; Chao, W.-r.; Pan, Ch.; Zhang, X.-K. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 1307. (f) Dawson, M. I.; Jong, L.; Hobbs, P. D.; Xiao, D.; Feng, K.-C.; Chao, W.-r.; Pan, Ch.; Fontana, J. A.; Zhang, X.-K. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 1311. (g) Yoshimura, K.; Kikuchi, K.; Hibi, S.; Tagami, K.; Satoh, T.; Ishibahi, A.; Tai, K.; Hida, T.; Tokuhara, N.; Nagai, M. *J. Med. Chem.* **2000**, *43*, 2929. (h) Kikuchi, K.; Hibi, S.; Yoshimura, K.; Tai, K.; Hida, T.; Tokuhara, N.; Yamauchi, T. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 619.
6. Shudo, K.; Kagechika, H. In *Chemistry and Biology of Synthetic Retinoids*; Dawson, M. I., Okamura, W. H., Eds.; CRC: Boca Raton, FL, 1990; Chapter 12, p 275.
7. Zhang, X.-K.; Lehmann, J.; Hoffmann, B.; Dawson, M. I.; Cameron, J.; Granpuer, G.; Hermann, T.; Pfahl, M. *Nature* **1992**, *358*, 587.
8. (a) Zhang, L.; Nadzan, A. M.; Heyman, R. A.; Love, D.; Mais, D. E.; Croston, G. E.; Lamph, W. L.; Boehm, M. F. *J. Med. Chem.* **1996**, *39*, 2659. (b) Bennani, Y. L.; Marron, K. S.; Mais, D. E.; Flatten, K.; Nadzan, A. M.; Boehm, M. F. *J. Med. Chem.* **1998**, *41*, 543.
9. Boehm, M. F.; Zhang, L.; Zhi, L.; McClurg, M. R.; Berger, E.; Wagoner, M.; Mais, D. E.; Suto, C. M.; Davies, P. J. A.; Heyman, R. A.; Nadzan, A. M. *J. Med. Chem.* **1995**, *38*, 3146.
10. Yokokawa, F.; Hamada, Y.; Shioiri, T. *Tetrahedron Lett.* **1993**, *34*, 6559.
11. (a) For reviews, see: Stille, J. K. *Pure Appl. Chem.* **1985**, *57*, 1771. (b) Stille, J. K. *Angew. Chem., Int. Ed. Engl.* **1986**, *25*, 508. (c) Mitchell, T. N. *Synthesis* **1992**, 803. (d) Sato, T. In *Comprehensive Organometallic Chemistry II*; Abel, E. W.; Stone, F. G. A.; Wilkinson, G., Eds.; Elsevier: Oxford, 1995; Vol. 11, Chapter 8, p 355. (e) Farina, V.; Roth, G. P. In *Advances in Metal-Organic Chemistry*; Liebeskind, L. S., Ed.; JAI: New York, 1996; Vol. 5, p 1. (f) Farina, V. *Pure Appl. Chem.* **1996**, *68*, 73. (g) Farina, V.; Krishnamurthy, V.; Scott, W. J. *The Stille Reaction. Org. React.* **1997**, *50*, 1. (h) Stanforth, S. P. *Tetrahedron* **1998**, *54*, 263. (i) Mitchell, N. T. In *Metal-Catalyzed Cross-Coupling Reactions*; Diederich, F., Stang, P. J., Eds; Wiley-VCH: New York, 1998; Chapter 4, p 167. (j) Farina, V.; Krishnamurthy, V. *The Stille Reaction*; Wiley: New York, 1999. (k) Duncton, M. A. J.; Pattenden, G. *J. Chem. Soc., Perkin Trans. 1* **1999**, 1235.
12. Negishi, E.; King, A. O.; Klima, W. L.; Patterson, W.; Silveira, A. *J. Org. Chem.* **1980**, *45*, 2526.
13. (a) Kamiya, N.; Chikami, Y.; Ishii, Y. *Synlett* **1990**, 675. (b) Balme, G.; Bouyssi, D. *Tetrahedron* **1994**, *50*, 403. (c) For a mechanism of this transformation, see: Luo, F.-T. *J. Org. Chem.* **1998**, *63*, 5656.
14. Farina, V.; Krishnan, B. *J. Am. Chem. Soc.* **1991**, *113*, 9585.
15. Boehm, M. F.; McClurgh, M. M.; Pathirana, C.; Mangelsdorf, D.; White, S. K.; Hebert, J.; Winn, D.; Goldman, M. E.; Heyman, R. A. *J. Med. Chem.* **1994**, *37*, 408.
16. Briefly, COS-7 cells were transfected with a pSG-derived expression vector encoding for human RXRα using the polybrene technique. Cells were lysed, and the nuclei were recovered by centrifugation. For competition binding assays, nuclear extracts were incubated with $[^3H]$ 9-*cis*-retinoic acid (50 nM) as the radioligand and various concentrations of the analogues. Separation of free and bound ligand was performed by using an hydroxylapatite gel. Dissociation constants values were determined by non-linear regression analysis using the Origin software (Microcalc Software Inc.). See: Martin, B.; Bernardon, J. M.; Cavey, M. T.; Bernard, B.; Carlavan, I.; Charpentier, B.; Pilgrim, W. R.; Shroot, B.; Reichert, U. *Skin Pharmacol.* **1992**, *5*, 57.
17. InsightII, Builder and Biopolymer are trademarks of MSI, Inc. San Diego CA, USA. The unsaturated side chain starting at atom 4′ and ending into the carboxylate group superimposes well with a similar chain that starts at atom C9 of the 9-*cis*-retinoic acid **2**. The RMSD betwen both chains is only 0.21 Å. To generate a set of conformers we fixed this fragment in the same conformation of the 9-*cis*-retinoic acid **2** starting at C6, and then we proceeded to run a 0.1 nanosecond (ns) molecular dynamics (MD) simulation of ligand in vacuum at 300 K. From this simulation we selected a set of 100 conformations equally spaced in time.
18. Egea, P. F.; Mitschler, A.; Rochel, N.; Ruff, M.; Chambon, P.; Moras, D. *EMBO J.* **2000**, *19*, 2592.
19. Brooks, B. R.; Bruccoleri, R. E.; Olafson, B. D.; States, D. J.; Swaminathan, S.; Karplus, M. *J. Comput. Chem.* **1983**, *4*, 187.
20. Choi, C. H.; Kertesz, M. *J. Phys. Chem. A* **1997**, *101*, 3823.