Pergamon

*Bioorganic & Medicinal Chemistry Letters*, Vol. 7, No. 5, pp. 561-566, 1997

0960-894X/97 $17.00 + 0.00

PII: S0960-894X(97)00067-X

# NOVEL SMALL MOLECULE NEUROTENSIN ANTAGONISTS: 3-(1,5-DIARYL-1,5-DIOXOPENTAN-3-YL)BENZOIC ACIDS

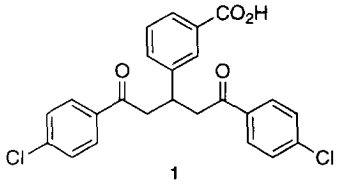
S. J. Johnson,* H. C. Akunne, T. G. Heffner, S. R. Kesten, T. A. Pugsley, L. D. Wise, and D. J. Wustrow

*Parke-Davis Pharmaceutical Research, Division of Warner-Lambert Company, 2800 Plymouth Rd., Ann Arbor, Michigan 48105.*

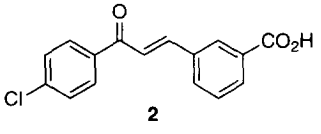
**Abstract:** Screening in a neurotensin (NT) receptor binding assay resulted in the discovery of compound **1** with NT receptor binding affinity of 450 nM. SAR studies that varied substituents on each aromatic ring and the linking 1,5-pentanedione chain lead to compound **39** with 42 nM affinity. The more potent compounds were shown to be NT antagonists by their ability to inhibit NT induced calcium mobilization in HT29 cells.


The neuropeptide neurotensin (NT) exerts regulatory hormone actions throughout the central nervous system[1] (CNS) and gastro-intestinal tract.[2] In the CNS, NT has a regulatory effect on dopaminergic neurotransmission.[3,4] NT has been shown to increase the dopamine content of the rat brain[5] and to increase the firing rate of dopaminergic neurons in certain brain regions.[6] The treatment of schizophrenia with neuroleptic drugs is believed to produce a reduction in the activity of dopaminergic neuronal pathways in the CNS. The possibility that NT antagonists could modulate the activity of dopaminergic systems offers an approach to the development of novel antipsychotic drugs with a new mechanism of action.[7] NT antagonists, such as SR 48962, have also been proposed as diuretic drugs[8] and for the treatment of gastric disorders.[9]

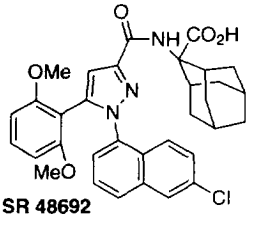
The Parke-Davis compound bank was screened in a NT receptor binding assay[10] using an immature mouse brain preparation. One of the more interesting hits, the acid **1** with 450 nM NT binding affinity, was the bisadduct from the aldol condensation of 4-chloroacetophenone with 3-formylbenzoic acid.[11] This compound's discovery was rather fortunate since it was originally prepared when the chalcone **2** had been the target. The chalcone **2** did not have any significant NT binding nor did any of its analogs used as intermediates in this project. The structure **1** possesses three aromatic rings separated along a 1,5-pentanedione chain. To investigate the SAR, we made variations with the working assumption that contributions to the NT binding by each ring would be independent. We also investigated several changes to the 1,5-pentanedione linker.



**1**



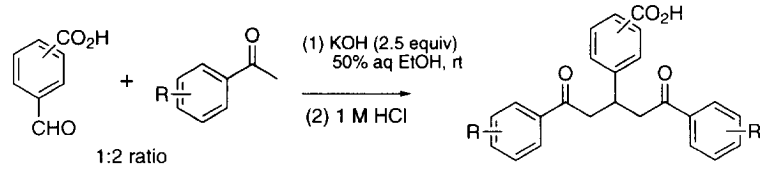
**2**



**SR 48692**

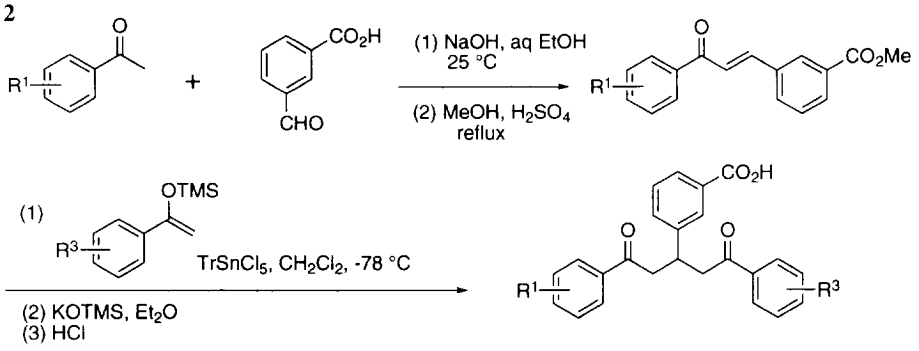
In most cases, the synthesis of symmetrical analogs of **1** was achieved using the simple aqueous base catalyzed aldol condensation and Michael addition of two equivalents of an acetophenone with a formylbenzoic acid (Scheme 1). The common starting material, 3-formylbenzoic acid, was best prepared by oxidation of isophthalaldehyde with one equivalent of iodine in aqueous NaOH/acetonitrile.[12]

**Scheme 1**



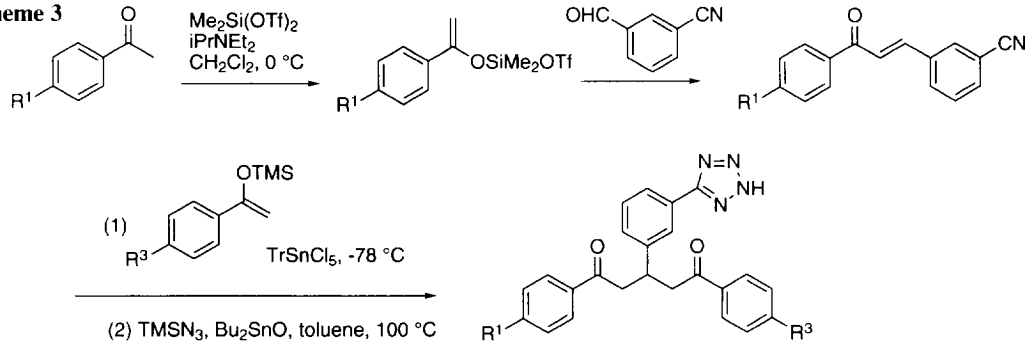

To prepare unsymmetrical analogs it was necessary to conduct the conjugate addition of the second acetophenone under nonequilibrating conditions[13] (Scheme 2). The mild ester hydrolysis with KOTMS[14] was necessary to avoid scrambling of $R^1$ and $R^3$ by retro-addition equilibration.

**Scheme 2**

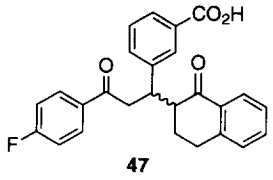



In cases where the substituents would not withstand aqueous base, such as the cyanide intermediate for the tetrazole synthesis[15] (Scheme 3), the aldol condensation was catalyzed by in situ generation of an activated silyl-enol ether.[16]

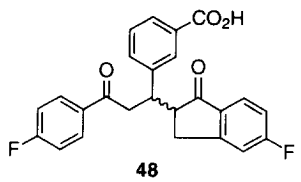
**Scheme 3**



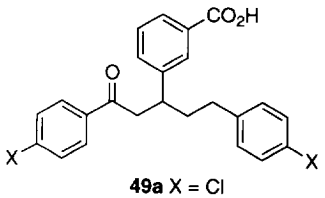
Several compound classes with changes to the 1,5-pentanedione were prepared by literature methods. Tetralone and indanone analogs (**47** and **48**) were prepared from the TMS-enol ethers by the route used in Scheme 2. The desoxy compounds (**49a** and **49b**) were produced by copper catalyzed conjugate addition of phenylethyl-samarium reagents to chalcones.[17,18] Cyclopropanes (**50**) were obtained by addition of a sulfur ylide to the chalcone.[19] Pyridine **51** was prepared from the pentanedione **1** by reaction with ammonia in acetic acid.[20]



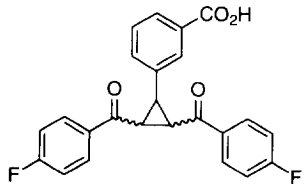

**47**



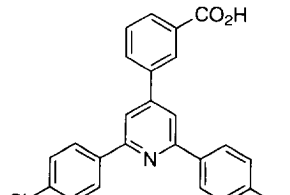

**48**




**49a** X = Cl
**49b** X = F

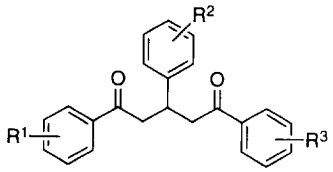



**50**




**51**

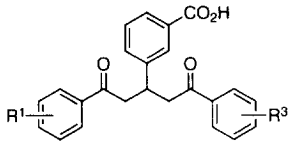
**Table 1: NT receptor binding in mouse brain tissue for symmetrical compounds where $R^1 = R^3$.**




| Compd | $R^1$ and $R^3$ | $R^2$ | NT, $K_i$ (nM) ± SEM | Compd | $R^1$ and $R^3$ | $R^2$ | NT, $K_i$ (nM) |
|---|---|---|---|---|---|---|---|
| **1** | 4-Cl | 3-$CO_2H$ | 450 ± 188 | **14** | 2,4-diCl | 3-$CO_2H$ | >10,000 |
| **3** | H | 3-$CO_2H$ | >10,000 | **15** | 2,4-diF | 3-$CO_2H$ | >10,000 |
| **4** | 2-Cl | 3-$CO_2H$ | >10,000 | **16** | 3,4-diF | 3-$CO_2H$ | >10,000 |
| **5** | 3-Cl | 3-$CO_2H$ | >10,000 | **17** | 4-F, 2-Me | 3-$CO_2H$ | >10,000 |
| **6** | 4-F | 3-$CO_2H$ | 302 ± 95 | **18** | 4-F, 3-Br | 3-$CO_2H$ | >10,000 |
| **7** | 4-Br | 3-$CO_2H$ | 1710 ± 200 | **19** | 4-F, 2-$CF_3$ | 3-$CO_2H$ | >10,000 |
| **8** | 4-I | 3-$CO_2H$ | >10,000 | **20** | 2,6-diOMe | 3-$CO_2H$ | >10,000 |
| **9** | 4-Me | 3-$CO_2H$ | >10,000 | **21** | 4-F | 2-$CO_2H$ | >10,000 |
| **10** | 4-$CF_3$ | 3-$CO_2H$ | >10,000 | **22** | 4-Cl | 4-$CO_2H$ | >10,000 |
| **11** | 3-$CF_3$ | 3-$CO_2H$ | >10,000 | **23** | 4-F | 3-tetrazole | 6320 |
| **12** | 4-tBu | 3-$CO_2H$ | >10,000 | **24** | 4-Cl | 3-tetrazole | 5900 |
| **13** | 3,4-diCl | 3-$CO_2H$ | >10,000 | **25** | 4-Cl | 3-CN | >10,000 |
| NT | | | 0.18 ± 0.06 | **26** | 4-F | 3-$CO_2Me$ | >10,000 |
| NT1 | | | 0.67 ± 0.03 | SR 48692 | | | 3.9 ± 0.1 |

The analogs prepared for this SAR study were evaluated in a [$^3$H]-NT binding assay in mouse brain.[10,21] Scatchard analysis of preliminary saturation experiments conducted in our laboratory showed [$^3$H]-NT binding was specific, reversible and saturable. [$^3$H]-NT binding was inhibited competitively by increasing concentrations of unlabelled neurotensin ligands. The results in Table 1 illustrate several aspects for the symmetrical adducts where $R^1 = R^3$. Firstly, only 4-chloro and 4-fluoro $R^1$ and $R^3$ substituents allowed significant NT binding (**1** and **6**). Secondly, only $R^2$ substituted as the 3-carboxylic acid retained substantial NT receptor binding activity (cf. **1** and **6** with **21-26**). Further generalization of SAR information obtained from these symmetrical adducts was limited because requirements for $R^1$ and $R^3$ may vary independently. Consequently, the symmetrical adducts provide information on a compromise between $R^1$ and $R^3$ requirements. Additional studies were conducted to determine preliminary SAR for variations of $R^1$ and $R^3$.

**Table 2: NT receptor binding for unsymmetrical compounds where $R^1$ does not equal $R^3$.**




| Compd | $R^1$ | $R^3$ | NT, $K_i$ (nM) ± SEM | Compd | $R^1$ | $R^3$ | NT, $K_i$ (nM) ± SEM |
|---|---|---|---|---|---|---|---|
| **27** | H | 4-F | 223 | **37** | 4-OMe | 4-Cl | 420 ± 90 |
| **28** | 4-Cl | 4-F | 440 | **38** | 2,4-diOMe | 4-Cl | 78 |
| **29** | 2-OMe | 4-F | 213 | **39** | 3,4-diOMe | 4-Cl | 42 |
| **30** | 4-OMe | 4-F | 329 ± 125 | **40** | 2,5-diOMe | 4-Cl | 260 ± 11 |
| **31** | 2,5-diOMe | 4-F | 331 | **41** | 2,6-diOMe | 4-Cl | 161 |
| **32** | 2,6-diOMe | 4-F | 392 | **42** | 3,4,5-triOMe | 4-Cl | 76 |
| **33** | 3,4-diOMe | 4-F | 153 | **43** | 2,3,4-triOMe | 4-Cl | 191 |
| **34** | 3,4,5-triOMe | 4-F | 204 | **44** | 4-Me | 4-Cl | 579 ± 30 |
| **35** | 2-OMe | 4-Cl | 158 | **45** | 2,6-diOMe | H | >10,000 |
| **36** | 3-OMe | 4-Cl | 136 | **46** | 2,5-diOMe | 4-CN | 1,120 |

The examples listed in Table 2 demonstrate that $R^1$ may be varied provided $R^3$ is kept as 4-F or 4-Cl. However, it should be noted that when $R^1$ does not equal $R^3$, the compounds are a mixture of enantiomers which differ essentially by reversal of the $R^1$ and $R^3$ substituents. We have been unable to separate these enantiomers employing chiral HPLC, which is not surprising given the distance of the differentiating groups $R^1$ and $R^3$ from the chiral center at C-3 in the pentanedione chain.

This preliminary SAR suggests that the NT receptor binding does differentiate between $R^1$ and $R^3$. The unsymmetrical compounds retained the requirement for $R^3$ to be 4-F or 4-Cl when $R^1$ was varied [cf. active **31** ($R^1$=2,5-diOMe, $R^3$=4-F) and **40** ($R^1$=2,5-diOMe, $R^3$=4-Cl) with almost inactive **46** ($R^1$=2,5-diOMe, $R^3$=4-CN); cf. active **32** ($R^1$=2,6-diOMe, $R^3$=4-F) and **41** ($R^1$=2,6-diOMe, $R^3$=4-Cl) with inactive **45** ($R^1$=2,6-diOMe, $R^3$=H)

and **20** ($R^1$=2,6-diOMe, $R^3$=2,6-diOMe); cf. active **27** ($R^1$=H, $R^3$=4-F) with inactive **3** ($R^1$=$R^3$=H ) and active **6** ($R^1$=$R^3$=4-F)]. The $R^1$ dimethoxyl, $R^3$ chlorine substituted compounds (e.g., **38**, **39**, **40**, **41** and **42**) are superior to any of the symmetrical compounds despite the fact that the symmetrical dimethoxyl substituted compound **20** was inactive. Given this differentiation of requirements at $R^1$ and $R^3$, it is possible that the NT binding activity resides in one enantiomer but proof of this awaits a practical chiral resolution of these asymmetric compounds.

None of the analogs with variants of the pentanedione (**47-51**) displayed significant NT binding.

Selected compounds were further evaluated in an in vitro functional assay. Compounds **6** (PD 149598), **39** (PD 156425) and **42** (PD 156556) were able to inhibit the calcium mobilization induced by NT in HT29 cells[22,23] with $IC_{50}$'s of 2.2, 10 and 4.6 μM respectively, thus demonstrating that the compounds act as NT antagonists. They were however significantly less potent than the reference compound,[24] SR 48692 in this assay ($IC_{50}$ 0.16 μM).

None of these compounds were able to reverse the hypothermia induced in mice by the small peptide NT agonist NT1.[21,25,26] SR 48692 also failed to antagonize this NT1 effect.[28] Nor did the compounds have any effect on mouse or rat locomotor activity.[27]

This failure to produce distinct in vivo CNS effects at doses of up to 30 mg/Kg (ip) suggests that these compounds may not be able to inhibit the CNS action of NT. However, the possibility remains that this new class of NT antagonist has subtype selectivity that precludes activity in these tests, as is apparent for SR 48692.[29-31]

**References**


1. Uhl, G. R.; Kuhar, J. M.; Snyder, S. H. *Proc. Nat'l. Acad. Sci. USA* **1977**, *74*, 4059.
2. Carraway, R.; Kitabgi, R.; Leeman, S. E. *J. Biol. Chem.* **1978**, *253*, 7996.
3. Kalivas, P. *Brain Research Reviews* **1993**, *18*, 75.
4. Maffrand, J. P.; Gully, D.; Boigegrain, R.; Kitabgi, P.; Rostene, W.; Le Fur, G. *Drugs of the Future* **1993**, *18*, 1137.
5. Faggin, B. M.; Zubieta, J. K.; Rezvani, A. M.; Cubeddu, L. X. *Pharm. Exp. Ther.* **1990**, *252*, 817.
6. Shi, W. X.; Bunney, B. S. *Brain Res.* **1991**, *543*, 315.
7. Chakravarty, P. K.; Ransom, R. W. *Current Pharmaceutical Design* **1995**, *1*, 317.
8. Croci, T. ; Sanofi. 1995, European Patent Application EP 95-401833.
9. Croci, T.; Guidice, A.; Antonina, M.; Gully, D.; Le Fur, G. *Br. J. Pharmacol.* **1995**, *115*, 383.
10. Mazella, J.; Chabry, J.; Katabji, P.; Vincent J.-P. *J. Biol. Chem.* **1988**, *263*, 144.
11. Johnson, S. J.; Kesten, S. R.; Wise, L. D.; Wustrow, D. J. ; Warner-Lambert Co. US patent 95-5430047.
12. Yamada, S.; Morizono, D.; Yamamoto, K. *Tetrahedron Lett.* **1992**, *33*, 4329.
13. Kobayashi, S.; Murakami, M.; Mukaiyama, T. *Chem. Letters* **1985**, 953.
14. Laganis, E. D.; Chenard, B. L. *Tet. Lett.* **1984**, *25*, 5831.
15. Wittenberger, S.; Donner, B. D. *J. Org. Chem.* **1993**, *58*, 4139.

16. Kobayashi, S.; Nishio, K. *J. Org. Chem.* **1993**, *58*, 2647.
17. Wipf, P.; Venkatraman, S. *J. Org. Chem.* **1993**, *58*, 3455.
18. Van de Weghe, P.; Collin, J. *Tet. Lett.* **1993**, *34*, 3881.
19. Tewari, R. S.; Awasthi, A. K. *Indian J. Chem.* **1980**, *19*, 155.
20. Brannock, K. C.; Davis, H. E. *J. Org. Chem.* **1966**, *31*, 980.
21. Akunne, H. C.; DeMattos, S. B.; Whetzel, S. Z.; Wustrow, D. J.; Davis, D. M.; Wise, L. D.; Cody, W. L.; Pugsley, T. A.; Heffner, T. G. *Biochem. Pharm.* **1995**, *49*, 1147.
22. Turner, J.; James-Kracke, M. R.; Camden, J. M. *J. Pharm. Exp. Ther.* **1990**, *253*, 1049.
23. Bozou, J. C.; Amis, S.; Vincent, J.-P.; Katabgi, P. *Mol. Pharmacol.* **1986**, *1986*, 489.
24. Nisato, D.; Guiraudou, P.; Barthelemy, G.; Gully, D.; Le Fur, G. *Fr. Life Sci.* **1994**, *54*, 95.
25. Heyl, D. L.; Sefler, A. M.; He, J. X.; Sawyer, T. K.; Wustrow, D. J.; Akunne, H. C.; Davis, M. D.; Pugsley, T. A.; Heffner, T. G.; Corbin, A. E.; Cody, W. L. *Int. J. Protein Res.* **1994**, *44*, 233.
26. Machida, R.; Tokumura, T.; Sasaki, A.; Tsuchiya, Y.; Abe, K. *J. Chromatogr.* **1990**, *534*, 190.
27. Heffner, T.; Downs, D.; Meltzer, L.; Wiley, J.; Williams, A. *J. Pharm. Biochem. Behavior* **1969**, *261*, 105.
28. Pugsley, T. A.; Akunne, H. C.; Whetzel, S. Z.; Demattos, S.; Corbin, A. E.; Wiley, J. N.; Wustrow, D. J.; Wise, L. D.; Heffner, T. G. *Peptides* **1995**, *16*, 37.
29. Dubuc, I.; Costentin, J.; Terranova, J. P.; Barnouin, M. C.; Soubrie, P.; Le Fur, G.; Rostene, W.; Kitabgi, P. *Br. J. Pharmacol.* **1994**, *112*, 352.
30. Mazella, J.; Botto, J.-M.; Guillemare, E.; Coppala, T.; Sarret, P.; Vincent, J.-P. *J. Neuroscience* **1996**, *16*, 5613.
31. Chalon, P.; Vita, N.; Kaghad, M.; Guillemot, M.; Bonnin, J.; Delpech, B.; Le Fur, G.; Ferrara, P.; Caput, D. *FEBS Lett.* **1996**, *386*, 91.

(*Received in USA* 7 *November* 1996; *accepted* 23 *January* 1997)