Pergamon

Bioorganic & Medicinal Chemistry Letters, Vol. 4, No. 9, pp. 1167-1170, 1994

0960-894X/94 $6.00+0.00

0960-894X(94)E0121-T

DERIVATIVES OF D-PENICILLAMINE AS POTENTIAL ANTIARTHRITIC AGENTS

Marvin F. Reich*, Bernard D. Johnson, Robert H. Lenhard and J. William Hanifin

Medical Research Division
American Cyanamid Company
Pearl River, New York 10965

Abstract: A variety of synthetic molecules based on D-penicillamine was examined in the established type II collagen arthritis model in rats.

D-(-)-Penicillamine (1) is a clinically useful antiarthritic agent belonging to a small group of such drugs which are believed to alter the course of this disease, rather than alleviate its symptoms.[1] They differ from the classical non-steroidal antiinflammatory drugs by not inhibiting arachidonic acid cyclooxygenase and in having a lag period of several months before beneficial effects are felt. Penicillamine is not consistently active in the bioassays commonly used for arthritis-carrageenin edema, UV erythema and adjuvant arthritis.[2]

An arthritis-like disease having similarities to the human condition has been induced in rats by injection of type II collagen.[3] We have adapted this animal model to screen for antiarthritic drugs, with x-rays being used to examine several parameters of joint destruction.[4] Other research groups have subsequently utilized this bioassay in their testing programs,[5] but most did not use joint x-rays.

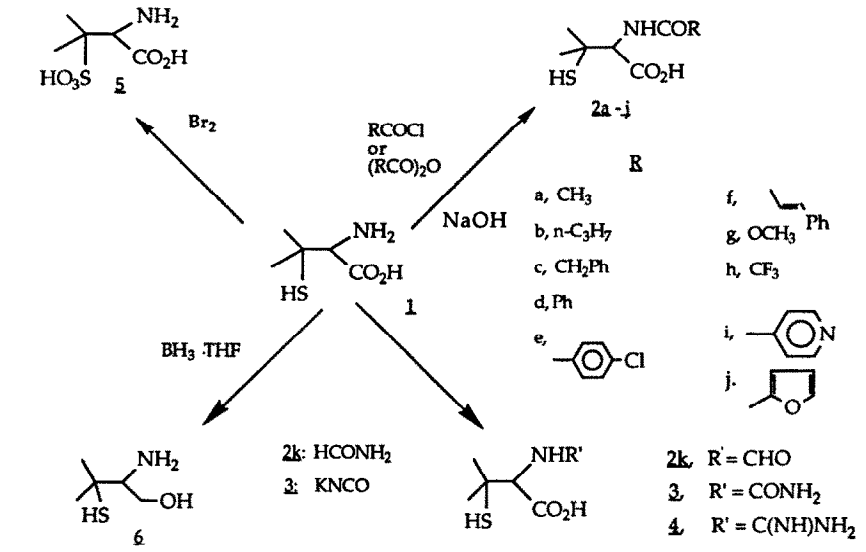
D-Penicillamine, but not the L-enantiomer, was initially found to have activity in our assay. A variety of analogs was then synthesized in which the chemical structure of D-penicillamine was systematically varied.

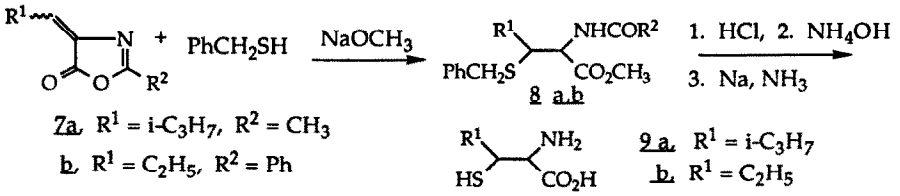
A series of N-acylated penicillamine derivatives (2a-j) was prepared by reaction of 1 with an acid chloride, anhydride, or chloroformate ester in the presence of base. The N-formyl compound (2k) was prepared by heating 1 in formamide.[6] Urea 3 was obtained with aq KNCO.[7] Preparation of the guanidine 4 required protection of the penicillamine sulfhydryl as its p-methoxybenzyl thioether, reaction with O-methylisourea hydrogen sulfate and deprotection (Hg^{++})[8].

Treatment of 2a with $NaOCH_3/CH_3I$ followed by deacylation (HCl,Δ) gave S-methylpenicillamine. The sulfhydryl group was oxidized to the sulfonic acid 5 with aq Br_2.[6] Penicillamine was converted to amino alcohol 6 with BH_3·THF.[9] Structures 2-6 incorporated the D-penicillamine stereochemistry.

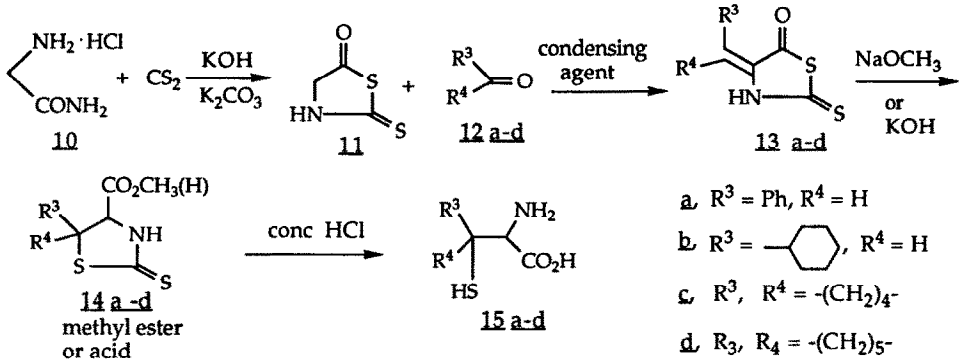
Racemic analogs of penicillamine containing modified alkyl groups were also synthesized. The oxazolones 7 were prepared by standard procedures[6] and then reacted with benzyl mercaptan to give the protected intermediates 8. Hydrolysis and debenzylation yielded the desired β–alkylcysteines (9).

Additional racemic modified cysteines were prepared as shown.[10] Glycinamide·HCl (10) was cyclized to 11 with CS_2 and base. Further reaction with a carbonyl compound (12) in the presence of HOAc, or better, $PhCH_2NH_2$[10c], as condensing agent yielded 13. Treatment of the latter with $NaOCH_3$ or KOH in CH_3OH gave rearrangement to 14 methyl ester or acid, respectively. Hydrolysis of 14 acid under forcing conditions led to 15.[11]

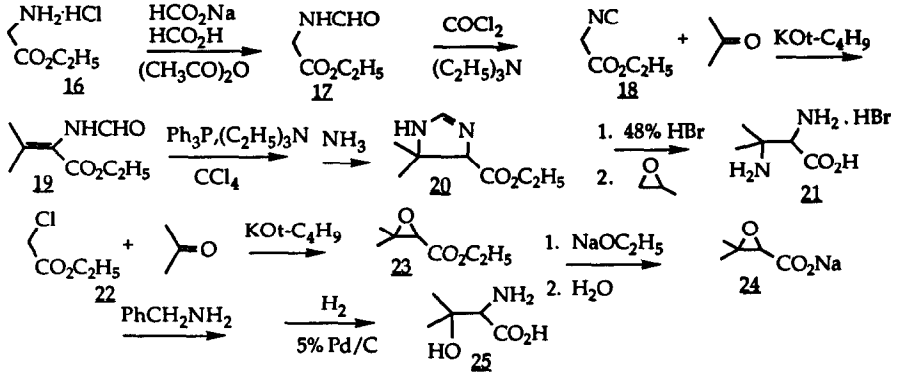




The sulfhydryl group of penicillamine is probably essential for its biological activity and may be the cause of its toxicity. Racemic compounds 21 and 25 in which the sulfhydryl group was replaced by other functionality were prepared. Glycine ethyl ester·HCl (16) was formylated[12] and dehydrated to yield 18.[13] Condensation with acetone gave 19.[14]



Dehydration, cyclization, acid hydrolysis and partial neutralization produced 21.[15] Glycidic ester condensation of acetone and ethyl chloroacetate gave 23. After careful saponification to 24, ring opening with $PhCH_2NH_2$ occurred on the α-carbon. Hydrogenolysis produced β-hydroxyvaline (25), the hydroxy analog of penicillamine.[16]



The established type II collagen arthritis assay was performed as previously described.[4] There were 3-18 rats treated with each compound at dosages up to 200 mg/kg, p.o. and the data were pooled. Arthritic paws were examined radiographically and scores were given for bone erosions and cartilage space (parameters related to joint destruction). Under these conditions, only D-penicillamine consistently showed improvement over control animals. Although the compounds were inactive, this is the first time that penicillamine analogs were examined in an assay of this type. D-and L-penicillamine could be distinguished; the latter being inactive. In control experiments, the clinically useful antiarthritic drugs indomethacin, phenylbutazone and prednisolone were active (Table 1). The latter three agents were found to decrease inflammation as measured by paw diameters, while D-penicillamine had no effect. This observation serves to distinguish these two classes of drugs.

Table 1. Examination of the Paws of Collagen Arthritic Rats[a]

Drug(dose,mg/kg)	Erosions	Cartilage Space	Mean Paw Diameter(mm)
Arthritis Control (0)	3.76 ± 0.09	3.6 ± 0.1	9.0 ± 0.12
D-Penicillamine (200)	2.83 ± 0.36	2.72 ± 0.36	8.89 ± 0.15
Indomethacin (2.0)	2.94 ± 0.33	2.94 ± 0.26	8.0 ± 0.11
Phenylbutazone (150.0)	3.33 ± 0.27)	2.86 ± 0.39	8.42 ± 0.21
Prednisolone (10.0)	3.50 ± 0.14	3.27 ± 0.16	7.92 ± 0.12

[a] Determinations were performed 42 days after initial immunization with type II collagen. Erosions of the tarsal bones and cartilage space narrowing were measured radiographically. Grades of 0-4 were used, where O is a normal animal and 4 is a severely diseased one. Paw diameters were measured with a micrometer caliper (normal rats = 7.04 ± 0.06 mm). An average of 18 rats was used for each drug. Details are given in reference 4.

References and Notes

1. (a) Kelley, W. N.; Harris, E. D., Jr.; Ruddy, S.; Sledge, C. B., Eds., "Textbook of Rheumatology", V. 1, 2nd ed.; W. B. Saunders Co., Philadelphia, PA, **1985**. (b) Huskisson, E. C., Ed., "Anti-Rheumatic Drugs", Praeger Publishers, East Sussex, UK, **1983**. (c) Stecher, V. J.; Carlson, J. A. *Ann. Rept. Med. Chem.* **1983**, 18, 171.
2. (a) Dunn, C. J.; Prouteau, M.; Delahaye, M.; Purcell, T.; Branceni, D. *Agents - Actions* **1984**, 14, 269. (b) Toivonen, M.L.; Tokola, O.; Vapaatalo, H. *Methods Find. Exp. Clin. Pharmacol.* **1982**, 4, 359. (c) Capasso, F.; Mascolo, N.; Autore, G. *Pharmacol. Res. Commun.* **1981**, 13, 385 and references therein. (d) Arrigoni-Martelli, E. *Agents - Actions* **1979**, Suppl. 5, 73.
3. Trentham, D. E.; Townes, A. S.; Kang, A. H. *J. Exp. Med.* **1977**, 146, 857 and subsequent papers.
4. (a) Kerwar, S. S.; Sloboda, A. E.; Birnbaum, J. E.; Oronsky, A. L. *J. Rheum.* **1981**, 8, Suppl. 7, 84. (b) Sloboda, A. E.; Birnbaum, J. E.; Oronsky, A. L.; Kerwar, S. S. *Arthritis and Rheumatism* **1981**, 24, 616.
5. (a) Lewis, A. J.; Carlson, R. P.; Chang, J.; DeLustro, F. *Agents - Actions* **1984**, 14, 707. (b) Probert, A. W., Jr.; Schrier, D. J.; Gilbertsen, R. B. *Arch. Int. Pharmacodyn. Ther.* **1984**, 269, 167. (c) Jones, S. A.; Kennedy, A. J.; Roberts, J. A. *Agents - Actions* **1982**, 12, 650. (d) Phadke, K.; Carroll, J.; Nanda, S. *Clin. Exp. Immunol.* **1982**, 47, 579.
6. Clarke, H. T.; Johnson, J. R.; Robinson, R., Eds., "The Chemistry of Penicillin", Princeton University Press, Princeton, NJ, **1949**, p. 455.
7. Poupaert, J. H.; Mukarugambwa, S.; Dumont, P. *Eur. J. Med. Chem.-Chim. Ther.* **1980**, 15, 511.
8. (a) Akabori, S.; Sakakibara, S.; Shimonishi, Y.; Nobuhara, Y. *Bull. Chem. Soc. Jpn.* **1964**, 37, 433. (b) Rudinger, J.; Poduska, K.; Zaoral, M. *Coll. Czech. Chem. Commun.* **1960**, 25, 2022. (c) Nishimura, O.; Kitada, C.; Fujino, M. *Chem. Pharm. Bull.* **1978**, 26, 1576.
9. Yoon, M. N.; Pak, C. S.; Brown, H. C.; Krishnamurthy, S.; Stocky, T. P. *J. Org. Chem.* **1973**, 38, 2786.
10. (a) Sweetman, B. J.; Vestling, M. M.; Ticaric, S. T.; Kelly, P. L.; Field, L.; Merryman, P.; Jaffe, I. A. *J. Med. Chem.* **1971**, 14, 868. (b) Cook, A. H.; Pollock, J. R. A. *J. Chem. Soc.* **1949**, 3007. (c) The use of benzylamine as condensing agent was suggested by Dr. M. D. Bachi, Weizmann Institute of Science, Israel. (d) Chatterjee, R.; Cook, A. H., Heilbron, I.; Levy, A. L. *J. Chem. Soc.* **1948**, 1337.
11. Spectral data for new compounds. DCA indicates dicyclohexylamine salt. N-acylated penicillamine derivatives displayed resonances in the following ranges: δ1.30-1.55 (s or 2s, 6H, ß-CH_3's), 4.10-4.65 (d, J=11Hz, 1H, Hα), 6.55-9.55 (d, J = 11Hz, 1H, NHCO); for dicyclohexylamine salts, 1.50-1.65 (m, 20H, CH_2's) 2.85-3.00 (s, br, 2H, CH). Spectral data unique to specific compounds follow (DMSO-d_6 unless otherwise noted). 2b, 0.875 (t, J = 8Hz, 3H), 1.60 (m, 2H), 2.20 (t, J = 9Hz, 2H), 2.88 (s, 1H), 12.8 (s, br, 1H); IR 1620, 1715 cm^{-1}. 2c $CDCl_3$ DCA 3.59 (s, 2H), 7.30 (s, 5H); IR 1635, 1663 cm^{-1}. 2d DCA 7.70 (m,6H); IR 1630, 1660 cm^{-1}. 2e DCA 7.50 (d, J=11 Hz, 2H), 7.85 (d, J = 11 Hz, 2H); IR 1635, 1655 cm^{-1}. 2f DCA 7.40 (m, 6H), 7.90 (d, J = 11 Hz, 1H); IR 1620, 1660 cm^{-1}. 2g 3.59 (s, 3H); IR 1720 cm^{-1}. 2h IR 1695, 1740 cm^{-1}. 2i 7.80 (d, J = 7 Hz, 2H), 8.80 (m, 2H); IR 1660, 1720 cm^{-1}. 2j 6.65 (m, 1H), 7.30 (d, J = 6 Hz, 1H), 8.40 (m, 2H); IR 1635, 1735 cm^{-1}. 6 $CDCl_3$ 1.40 (s, 6H), 2.05 (s, 4H), 2.70 (dd, J = 9, 5 Hz, 1H), 3.34 (AB, J = 10, 13 Hz, 1H), 3.80 (dd, J = 14, 5 Hz, 1H). 9a CF_3CO_2D, 1.30 (s, 6H), 2.10 (s, br, 1H), 3.60 (s, br, 1H), 4.80 (s, br, 1H). 9b CF_3CO_2D, 1.20 (m, 3H), 1.90 (m, 2H), 3.55 (m, 1H), 4.60 (m, 1H), 15b 1.30 (m, 11H), 3.65 (s, br, 1H), 5.70 (s, br, 1H).
12. (a) Jones, R. G. *J. Am. Chem. Soc.* **1949**, 71, 644. (b) Physical Properties: Martin, P. K.; Matthews, H. R.; Rapoport, H.; Thyagarajan, G. *J. Org. Chem.* **1968**, 33, 3758.
13. Ugi, I.; Fetzer, U.; Eholzer, U.; Knupfer, H.; Offermann, K. *Angew. Chem. (Int. Ed. Engl.)* **1965**, 4, 472.
14. Schöllkopf, U.; Meyer, R. *Ann. Chem.* **1977**, 1174.
15 Meyer, R.; Schöllkopf, U.; Böhme, P. ibid 1183.
16. (a) Johnson, W. S.; Belew, J. S.; Chinn, L. J.; Hunt, R. H. *J. Am. Chem. Soc.* **1953**, 75, 4995. (b) Oh-Hashi, J-i; Harada, K. *Bull. Chem. Soc. Jpn.* **1966**, 39, 2287. (c) Kaneko, T.; Inui, T. *Nippon Kagaku Zasshi* **1961**, 82, 743; *Chem. Abs.* **1963**, 59, 742e.

(*Received in USA* 17 *August* 1993; *accepted* 29 *March* 1994)