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DUAL METALLOPROTEASE INHIBITORS.V. UTILIZATION OF BICYCLIC AZEPINONETHIAZOLIDINES AND AZEPINONETETRAHYDROTHIAZINES IN CONSTRAINED PEPTIDOMIMETICS OF MERCAPTOACYL DIPEPTIDES

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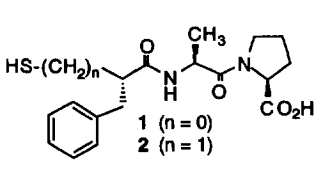
Abstract: Incorporation of mercaptoacetyl or mercaptopropanoyl groups into azepinonethiazolidine and azepinonetetrahydrothiazine carboxylic acids provided conformationally restricted peptidomimetics of Ala-Pro exhibiting potent dual activity *in vitro* versus ACE and NEP.

The search for dual-acting inhibitors of the zinc metalloproteases angiotensin converting enzyme (ACE) and neutral endopeptidase (NEP) as a potential new treatment for hypertension and congestive heart failure is actively being pursued by a number of groups.[1] ACE is the dipeptidyl carboxypeptidase that catalyzes the cleavage of the decapeptide angiotensin I (AI) to the vasoconstrictor octapeptide angiotensin II (AII). NEP is the enzyme that catalyzes the cleavage and inactivation of atrial naturetic peptide (ANP), a 28-amino acid peptidic hormone that promotes diuresis, natriuresis, and vasodilatation.[1a,1c,2] Co-administration of selective inhibitors of ACE and NEP in models of hypertension[3] and heart failure[4] have shown beneficial synergistic effects in comparison to administration of selective ACE or NEP inhibitors alone.



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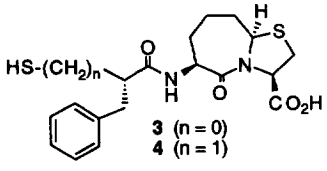

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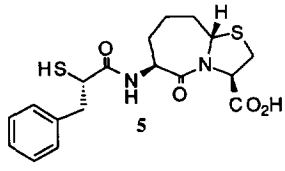

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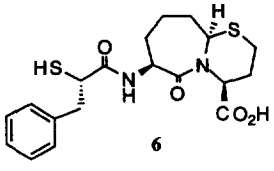

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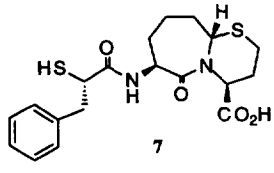

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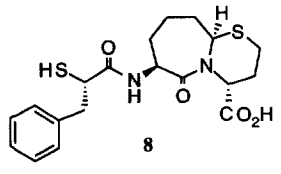

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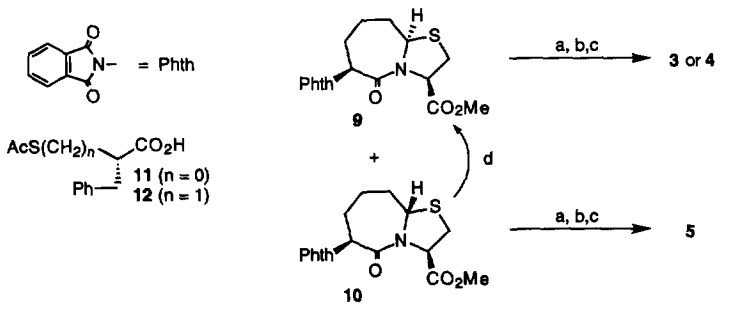
Recent reports from our laboratories have described the generation of dual ACE/NEP inhibitors utilizing constrained peptidomimetics of the Ala-Pro portion of mercaptoacyl dipeptides **1** and **2**.[5] Patchett and Wyvratt have described the synthesis of bicyclic azepinonethiazolidines **9** and **10** and shown that their substitutions for the Ala-Pro fragment in enalapril led to potent inhibitors of ACE *in vitro*.[6a,b] Additionally, utilization of the

corresponding azepinonetetrahydrothiazine analogs of enalaprilat were claimed to result in effective inhibitors of ACE.[6c] Herein, we describe the synthesis and dual ACE/NEP activity of mercaptoacetyl (propanoyl) azepinonethiazolidines **3**, **4**, and **5** and mercaptoacetyl azepinonetetrahydrothiazines **6**, **7**, and **8**.

The synthesis of compounds **3-5** is shown in Scheme 1. Phthalimido protected compounds **9** and **10** were prepared as described in the patent literature.[6b] Removal of the phthalimido protecting group from either **9** or **10** gave the corresponding amine, which was acylated with either (S)-α-(acetylthio)-3-benzenepropanoic acid (**11**)[5c,8] or (S)-3-(acetylthio)-2-benzylpropionic acid (**12**)[9]. Subsequent saponification in the absence of oxygen followed by acidification provided the desired acid **3**, **4**, or **5**.

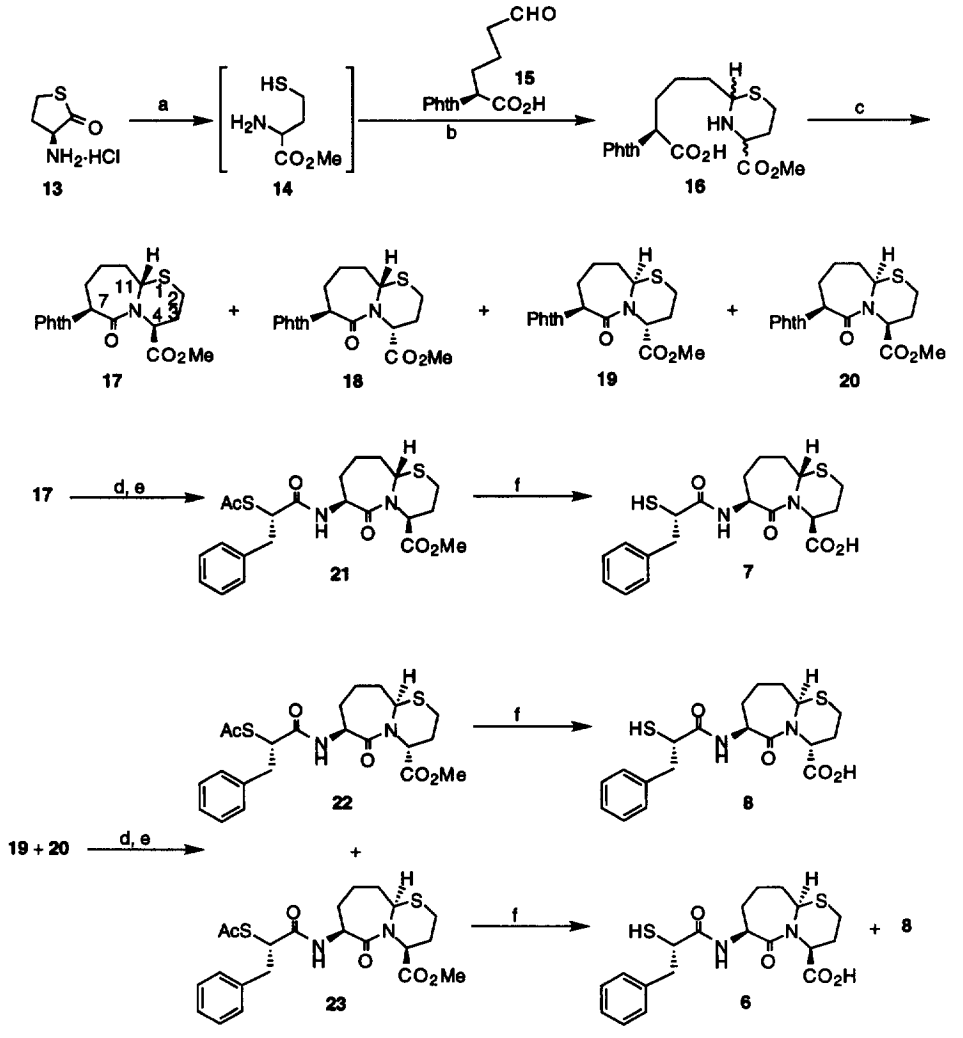
Scheme 1



a) H_2N-NH_2·H_2O, EtOH (95-100%); b) BOP reagent, Et_3N, CH_2Cl_2 (63-74%); c) aq. NaOH, MeOH then H^+ (54-80%); d) TsOH·H_2O, benzene, Δ, (81%).

Homochiral azepinonetetrahydrothiazines **6-8** were prepared by a procedure analogous to that described for generation of the azepinonethiazolidines and is outlined in Scheme 2. Attempted preparation of two tetrahydrothiazines **16** by sequential treatment of L-homocysteine thiolactone hydrochloride (**13**) with sodium methoxide (2 equivalents) and homochiral aldehyde **15**[6b] gave a complex mixture of isomers, presumably via racemic methyl homocysteine (**14**). Reaction of the mixture **16** with EEDQ provided the isomers **17** (25%), **18** (8%), **19** (21%), and **20** (1%).[10] Isomers **17**, **18**, and **19** were each found to equilibrate with acid (TsOH, refluxing benzene, 2-4 h) to give identical mixture of isomers **17**, **18**, **19**, and **20** in the respective ratio 10 : 30 : < 1 : 15.[11] In contrast, acid catalyzed isomerization of either azepinonethiazolidine **9** or **10** provided only compound **9**.[6b] Removal of the phthalimido protecting group from **17**, subsequent acylation with **11**, and final deprotection gave compound **7** whose structure and absolute configuration were confirmed by a single crystal X-ray analysis. Isomers **19** and **20** could not be separated by silica gel chromatography but were readily separable as their acylated products **22** and **23**. Initial treatment of a mixture (ca. 4 : 1) of isomers **19** and **20**, respectively, with hydrazine and subsequent acylation with **11** afforded **22** (50%) and **23** (15%) after chromatography. Deprotection of **22** provided compound **8** whose structure and absolute configuration were also confirmed by a single crystal X-ray analysis. Removal of the protecting groups from **23**, however, gave not only the expected acid **6** (40%) but also the related epimer **8** (30%), which were separable by chromatography on silica gel. Partial epimerization of the carboxyl group in **8** in an acetylation-deacetylation sequence provided **6** in 22% yield thus confirming that the stereoconfigurations at C-7 and C-11 in both acids are identical.[12]

Scheme 2



a, NaOMe (2 equiv.), MeOH; b, **15**, MeOH; c, EEDQ, THF (**17**, 25%; **18**, 8%; **19**, 21%; **20**, 1%; 3 steps); d, H_2N-NH_2-H_2O, MeOH, CH_2Cl_2; e, **11**, BOP reagent, Et_3N, CH_2Cl_2; f, aq. NaOH, MeOH then H^+ (**8**, 77% from **22**; **6**, 40% and **8**, 30% from **23**)

The *in vitro* NEP and ACE activities for compounds **3-8** and the ACE activity *in vivo* (intravenous administration (iv) in the AI challenged normotensive rat) for compounds **3** and **4** are shown in Table 1. In the azepinonethiazolidine series, greater ACE and NEP activity *in vitro* was seen for the mercaptopropanoyl compound **4** than the mercaptoacetyl analog **3**. *In vivo*, however, mercaptopropanoyl **4** was found to be 11 fold less active versus ACE than its mercaptoacetyl analog **3**. Superior ACE activity *in vivo* for mercaptoacetyl analogs as compared to their mercaptopropanoyl counterparts has also been demonstrated for other

conformationally restricted dual ACE/NEP inhibitors.[5c] Changes in the bridgehead configuration of conformationally restricted analogs **3** and **5** were found to effect the inhibitory potency versus NEP much more than ACE. A slight increase in activity versus ACE *in vitro* was seen for the 10 β-H analog **5** as compared to the 10 α-H compound **3**, whereas a 260 fold decrease in activity was observed versus NEP. In the azepinonetetrahydrothiazine series, 7,6-fused inhibitor **6** displayed somewhat weaker activity against both NEP and ACE *in vitro* compared to the corresponding 7,5-fused azepinonethiazolidine **3**. Changing the configuration at the ring juncture of **6** from α-H to β-H as in **7** produced a slight enhancement in activity versus ACE but only a 3 fold decrease in activity versus NEP. Alteration in configuration of the carboxyl group in **6** from β to α (i.e. compound **8**) resulted, as expected, in a substantial loss in activity against both ACE and NEP.

Table 1

Compound	NEP (I_{50}, nM)	ACE (I_{50}, nM)	[a]ACE ED_{50} (μM/kg, iv)
3	11.5	16	0.07
4	1.1	5.5	0.80
5	2820	11.5	NT
6	42	25	NT
7	133	11.6	NT
8	937	205	NT

[a]dose required to effect 50% inhibition of the AI induced pressor response;
NT = not tested

In conclusion, azepinonethiazolidines and azepinonetetrahydrothiazines were utilized as conformationally restricted surrogates of Ala-Pro in designing mercaptoacyl containing dual inhibitors of ACE and NEP. Compounds **3**, **4**, and **6** were found to have potent activity *in vitro* against both ACE and NEP. In the thiazolidine series, mercaptopropanoyl compound **4** exhibited greater activity than mercaptoacetyl analog **3** versus ACE *in vitro* but displayed appreciably less activity than **3** against ACE *in vivo*. In both series, NEP was found to be sensitive to changes in configuration at the bridgehead position, whereas ACE was relatively less effected.

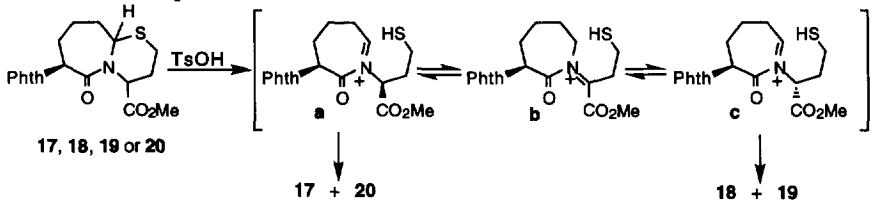
Acknowledgements

We are grateful for the technical assistance provided by Ms. Mary Giancarli and Mr. Hong Son Cheung. Appreciation is also expressed to Dr. Jack Gougoutas for helpful discussions in regard to the crystallographic analysis of compounds **7** and **8**.

References and Notes

1. (a) Roques, B. P. and Beaumont, A. *Trends Pharmacol. Sci.*, **1990**, *11*, 245. (b) Gros, C.; Noel, N.; Souque, A.; Schwartz, J. -C.; Danvy, D.; Plaquevent, J. -C.; Duhamel, L.; Duhamel, P.; Lecomte, J.-M.; Bralet, J. *Proc. Natl. Acad. Sci. USA* **1991**, *88*, 4210 . (c) Fournie-Zaluski, M. C. *C. R. Soc. Biol.*, **1992**,*186*, 612. (d) Roques, B.P.; *Biochem. Soc. Trans.* **1993**, *21*, 678. (e) Flynn, G. A.; Beight, D. W.; Mehdi, S.; Koehl, J. R.; Giroux, E. L.; French, J. F.; Hake, P. W.; Dage, R. C. *J. Med Chem.* **1993**, *36*, 2420. (f) Stanton, J. L.; Sperbeck, D. M.; Trapani, A. J.; Coric, P.; Turcaud, S.; Rousselet, N.; Gonzalez, W.; Barbe, B,; Pham, I.; Jullian, N.; Michel, J. -B.; Roques, B. P. *J. Med. Chem.* **1994**, *37*, 1070. (g) Fournie-Zaluski, M. -C.; Coric, P.; Turcaud, S.; Rousselet, N.; Gonzalez, W.; Barbe, B.; Pham, I.; Jullian, N.; Michel, J. B.; Roques, B. P. *J. Med. Chem.* **1994**, *37*, 1070-1083.
2. (a) Koehn, J. A.; Norman, J. A.; Jones, B. N.; LeSueur, L.; Sakane, Y. and Ghai, R. D. *J. Biol. Chem.* **1987**, *262*, 11623. (b) Stephenson, S. L. and Kenny, A. J. *Biochem. J.* **1987**, *243*, 183. (c) Schwartz, J. C.; Gros, C.; Lecomte, J. M.; Bralet, *J. Life Sci.* **1990**, *47*, 1279. (d) Brenner, B. M.; Ballerman, B. J.; Gunning, M. E.; Zeidel, M. L. *Physiol. Rev.* **1990**, *70*, 665. (e) Roques, B. P.; Noble, R.; Dauge, V.; Fournie-Zaluski, M.-C.; Beaumont, A. *Pharm. Rev.* **1993**, *45*, 87. (f) Wilkins, M. R.; Unwin, R. J.; Kenny, A. J. *Kidney Internat.* **1993**, *43*, 273. (g) Welches, W. R.; Brosnihan, K. B.; Ferrario, C.M. *Life Sci*, **1993**, *52*, 1461.
3. (a) Seymour, A. A.; Swerdel, J. N.; Abboa-Offei, B. J. *J. Cardiovasc. Pharrm.* **1991**, *17*, 456. (b) Krulan, C.; Ghai, R. D.; Lappe, R. W.; Webb, R. L. *FASEB J* **1993**, *7*, A247. (c) Pham, I.; Gonzalez, W.; Ell Amarani, A. -I. K.; Fournie-Zaluski, M. -C.; Philippe, M .; Loboulandine, I.; Roques, B. P.; Michel, J. -B. *J. Pharmacol. Exp. Ther.***1993**, *265*, 1339.
4. (a) Trippodo, N. C.; Fox, M. ; Nataajan, V.; Panchal,B. C.; Dorso, C. R.; Asaad, M. M. *J. Pharmacol. Exp. Ther.* **1993**, *267*, 108. (b) Seymour, A. A.; Asaad, M. M.; Lanoce, V. M.; Langen bacher, K. M.; Fennell, S. A.; Rogers, W. L. *J. Pharmacol. Exp. Ther.* **1993**, *266*, 872.
5. (a) Delaney, N. G.; Barrish, J. C.; Neubeck, R.; Natarajan, S.; Cohen, M.; Rovnyak, G. C.; Huber, G.; Murugesan, N.; Girotra, R.; Sieber-McMaster, E.; Robl, J. A.; Asaad, M. M.; Cheung, H. S.; Bird, J. E.; Waldron, T.; Petrillo, E. W. *Bioorg. Med. Chem. Letters* **1994**, *4*, 1783. (b) Robl, J. A.; Simpkins, L. M.; Sulsky, R.; Sieber-McMaster, E.; Stevenson, J.; Kelly, Y. F.; Sun, C.-Q.; Misra, R. N.; Ryono, D. E.; Asaad, M. M.; Bird, J. E.; Trippodo, N. C.; Karanewsky, D.S. *Bioorg. Med. Chem. Letters* **1994**, *4*, 1795. (c) Robl, J. A.; Simpkins, L. M.; Stevenson, J.; Sun, C.-Q.; Murugesan, N. Barrish, J. C.; Asaad, M. M.; Bird, J. E.; Schaeffer, T. R.; Trippodo, N. C.; Petrillo, E. W.; Karanewsky, D. S. *Bioorg. Med. Chem. Letters* **1994**, *4,* 1789. (d) Robl, J. A.; Sun, C.-Q.; Simpkins, L. M.; Ryono, D. E.; Barrish, J. C.; Karanewsky, D. S.; Asaad, M. M.; Schaeffer, T. R.; Trippodo, N. C. *Bioorg. Med. Chem. Letters* **1994**, *4*, 2055-2060. (e) Das, J.; Robl, J. A.; Reid, J. A.; Sun, C.-Q.; Misra, R. N.; Brown, B. R.; Ryono, D. E.; Asaad, M. M.; Bird, J. E.; Trippodo, N. C.; Petrillo, E. W.; Karanewsky, D. S. *Bioorg. Med. Chem. Letters* **1994**, *4*, 2193-2198.
6. (a) Wyvratt, M. J.; Patchett, A. A. *Med. Chem. Rev.* **1985**, *5*, 483. (b) *European Patent Application* 61187 (**1982**). (c) *South African Patent Application* 857527 (**1985**).

7. In recent patent applications we and workers from Marion Merrell Dow independently described the synthesis of compounds such as **3** and **5**. See (a) *European Patent Application* 599444A1 (**1994**). (b) World Patent Application 9410193 (**1994**).
8. Strijtveen, B.; Kellogg, R. M. *J. Org. Chem.* **1986**, *51*, 3664.
9. Giros, B.; Gros, C.; Schwartz, J. C. Danvy, D.; Plaquevent, J. C.; Duhamal. L.; Duhamel, P.; Vlaiculescu, A.; Constentin, J.; Lecomte, J. M. *J. Pharmacol. Exp. Ther.* **1987**, *243*, 666.
10. The structures of isomers **17** and **18** were assigned from NOE and COSY experiments in combination with molecular modelling studies (Macromodel). NOEs were not observed between methine protons at H-7 and H-11 in **17** and **18**, whereas NOEs were found between these protons in isomer **19** indicating a *trans* relationship for protons H-7 and H-11 in **17** and **18** and a *cis* relationship between H-7 and H-11 in **19**. Additionally, NOEs were found between protons H-4 and H-11 in **18** but not in **17** indicating a *cis* relationship between protons H-4 and H-11 in **18** and a *trans* relationship between protons H-4 and H-11 in **17**.
11. The interconversion of isomers **17-20** upon treatment with TsOH likely proceeds through an equilibrium mixture of iminium species **a**, **b**, and **c**.



12. Acetylation of **8** (Ac_2O, deoxygenated aq. dioxane, $KHCO_3$) and subsequent deacetylation (NH_4OH, aq. CH_3OH) afforded **6** (22%) along with recovered **8** (66%) after chromatography.

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