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Prolyl Endopeptidase Inhibitors. II. A Peptidyl α-Keto Thiazole Derivative

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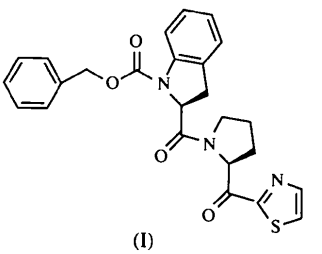
(*Received* 18 *April* 1994; *accepted* 16 *March* 1995)

Abstract

In benzyl 2-[2-(2-thiazoloyl)pyrrolidinoyl]indoline-1-carboxylate, $C_{25}H_{23}N_3O_4S$, the N-terminal urethane bond is *cis* and the proline amide bond is *trans*. The dipeptide adopts a polyproline II conformation and shows coplanarity of the ketone carbonyl group and the thiazole ring, with the carbonyl O atom *cis* with respect to the ring S atom.

Comment

The rational design and synthesis of protease inhibitors is an attractive field in medicinal and bioorganic chemistry (Rich, 1990). A general approach has been the replacement of the scissile amide unit by an electron-deficient carbonyl group (Wiley & Rich, 1993). Prolyl endopeptidase (PEP) (E.C. 3.4.21.26) is a serine protease that cleaves proline-containing peptides such as substance P, vasopressin and bradykinin (Welches, Brosnihan & Ferrario, 1993). It is thought that PEP inhibitors may improve learning and memory by prolonging the half-life of neuropeptides (Angelucci *et al.*, 1993). The peptidyl α-keto thiazole compound was found to be more potent as an inhibitor than both the α-keto ester and aldehyde derivatives (Tsutsumi *et al.*, 1994). The α-keto heterocyclic functional group may be an effective bioisostere of the α-keto ester moiety. The structure determination of the new α-keto thiazole inhibitor, (I), was undertaken as a step towards elucidating the inhibition mechanism.



(I)

The title dipeptide, with $\varphi_1 = -67(1)$, $\psi_1 = 143(9)$, $\varphi_2 = -74(1)$ and $\psi_2 = 150(1)°$ (IUPAC–IUB Commission on Biochemical Nomenclature, 1971), has a polyproline II conformation. Recent structure determinations of Src homology 3 (SH3) domains from PI3K, fyn and Grb2 complexed with proline-rich ligands indicate that SH3 domains recognize the polyproline II conformation (Lim, Richards & Fox, 1994; Feng, Chen, Yu, Simon & Schreiber, 1994). The present study may aid the design of small compounds interacting with SH3 domains. The N-terminal urethane bond is *cis* [O2—C9—N1—C8 −2(1)°]. Conformationally constrained amino acids like indolecarboxylic acid permit such a *cis* conformation (Magaard, Sanchez, Bean & Moore, 1993; Tsutsumi, Okonogi, Takeuchi & Kodama, 1995). The proline amide bond is *trans* [C8—C19—N2—C20 179(8)°]. The ketone carbonyl group and the thiazole ring are coplanar [O4—C21—C22—S1 −2(2) and N3—C22—C21—O4 173(1)°]. The carbonyl O atom of the ketone moiety and the S atom of the thiazole ring are *cis* with respect to each other.

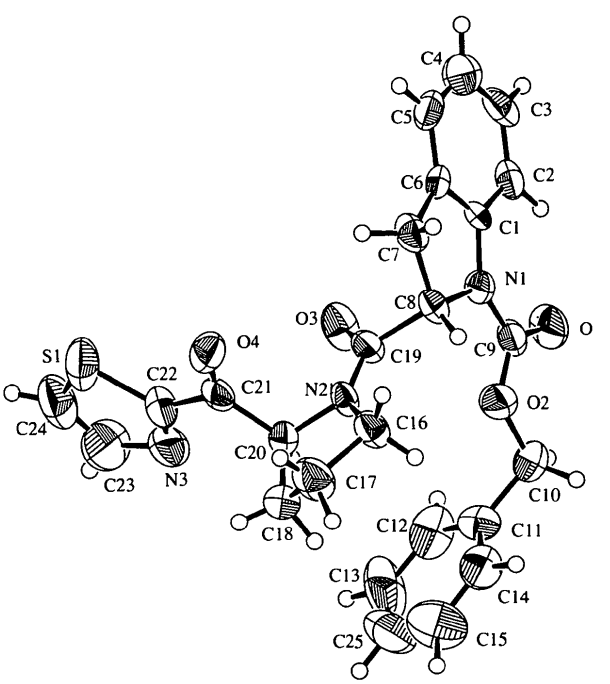


Fig. 1. The molecular structure of the title compound with the crystallographic numbering scheme (*ORTEPII*; Johnson, 1976). Displacement ellipsoids are shown at the 50% probability level and H atoms are drawn as spheres of arbitrary size.

Recently, Edwards *et al.* (1992) determined the X-ray crystal structure of the covalent complex between elastase and an α-keto benzoxazole inhibitor (Edwards, Wolanin, Andisik & Davis, 1995). We believe that peptidyl α-keto heterocyclic inhibitors which possess an N atom at the β position may also stabilize the hemiketal adduct through the formation of a hydrogen bond with the histidine of the active site, as in the elastase complex.

Experimental

The title compound was synthesized according to the method of Okonogi *et al.* (1993). Single crystals were grown from an ethyl acetate solution.

Crystal data

$C_{25}H_{23}N_3O_4S$
$M_r = 461.53$
Monoclinic
$P2_1$
$a = 8.819\,(1)$ Å
$b = 6.656\,(4)$ Å
$c = 20.006\,(4)$ Å
$\beta = 93.34\,(1)°$
$V = 1172.2\,(8)$ Å^3
$Z = 2$
$D_x = 1.307$ Mg m^{-3}
Cu $K\alpha$ radiation
$\lambda = 1.5418$ Å
Cell parameters from 22 reflections
$\theta = 10.45$–$20.67°$
$\mu = 1.487$ mm^{-1}
$T = 23.0$ K
Plate
0.2 × 0.06 × 0.04 mm
Colorless

Data collection

AFC-5*R* diffractometer
ω scans with profile analysis
Absorption correction: none
1976 measured reflections
1917 independent reflections
1912 observed reflections $[I > 0]$
$R_{int} = 6.40$
$\theta_{max} = 60°$
$h = -9 \rightarrow 9$
$k = 0 \rightarrow 7$
$l = 0 \rightarrow 14$
3 standard reflections monitored every 150 reflections
intensity decay: 0.20%

Refinement

Refinement on F
$R = 0.1940$
$wR = 0.075$
$S = 1.170$
1912 reflections
297 parameters
H-atom parameters not refined
$w = 1/\sigma^2(F_o)$
$(\Delta/\sigma)_{max} = 0.62$
$\Delta\rho_{max} = 0.53$ e Å^{-3}
$\Delta\rho_{min} = -0.72$ e Å^{-3}
Extinction correction: type 2 Gaussian isotropic (Zachariasen, 1967)
Extinction coefficient: 2.16×10^{-7}
Atomic scattering factors from *International Tables for X-ray Crystallography* (1974, Vol. IV)

Table 1. *Fractional atomic coordinates and equivalent isotropic displacement parameters* (Å^2)

$$U_{eq} = (1/3)\Sigma_i\Sigma_j U_{ij} a_i^* a_j^* \mathbf{a}_i.\mathbf{a}_j.$$

	x	y	z	U_{eq}
S1	0.8798 (5)	0.8099	0.4910 (2)	0.103 (1)
O1	0.5166 (8)	0.661 (2)	0.0604 (3)	0.091 (3)
O2	0.5869 (8)	0.411 (2)	0.1322 (3)	0.079 (3)
O3	0.7379 (10)	0.723 (1)	0.2477 (4)	0.082 (3)
O4	0.9870 (8)	0.569 (2)	0.3777 (3)	0.081 (3)
N1	0.7520 (10)	0.650 (2)	0.1109 (4)	0.060 (3)
N2	0.8100 (9)	0.405 (2)	0.2700 (4)	0.049 (3)
N3	0.629 (1)	0.674 (2)	0.4394 (5)	0.105 (5)
C1	0.815 (1)	0.826 (2)	0.0884 (4)	0.046 (3)
C2	0.747 (1)	0.970 (2)	0.0467 (5)	0.062 (4)
C3	0.835 (2)	1.135 (2)	0.0305 (5)	0.077 (5)
C4	0.983 (2)	1.148 (2)	0.0550 (6)	0.088 (5)
C5	1.049 (1)	1.007 (2)	0.0979 (5)	0.067 (4)
C6	0.961 (1)	0.843 (2)	0.1137 (4)	0.059 (4)
C7	1.006 (1)	0.666 (2)	0.1557 (4)	0.070 (4)
C8	0.857 (1)	0.542 (2)	0.1595 (4)	0.053 (3)
C9	0.610 (1)	0.580 (2)	0.0981 (5)	0.059 (4)
C10	0.445 (1)	0.314 (3)	0.1223 (6)	0.102 (5)
C11	0.402 (1)	0.225 (2)	0.1884 (6)	0.065 (4)
C12	0.329 (2)	0.333 (3)	0.2324 (8)	0.098 (6)
C13	0.295 (2)	0.245 (4)	0.2932 (10)	0.149 (10)
C14	0.444 (1)	0.033 (3)	0.2030 (7)	0.088 (6)
C15	0.408 (2)	−0.062 (3)	0.261 (1)	0.131 (8)
C16	0.870 (1)	0.207 (2)	0.2574 (5)	0.067 (4)
C17	0.886 (1)	0.107 (2)	0.3265 (6)	0.091 (5)
C18	0.758 (1)	0.204 (2)	0.3611 (5)	0.076 (5)
C19	0.790 (1)	0.568 (2)	0.2313 (5)	0.062 (4)
C20	0.751 (1)	0.420 (2)	0.3374 (5)	0.063 (4)
C21	0.852 (1)	0.552 (2)	0.3812 (5)	0.060 (4)
C22	0.776 (1)	0.665 (2)	0.4345 (5)	0.076 (4)
C23	0.596 (2)	0.798 (3)	0.4941 (8)	0.134 (7)
C24	0.717 (2)	0.883 (2)	0.5234 (6)	0.113 (7)
C25	0.338 (2)	0.051 (4)	0.304 (1)	0.132 (10)

The intensities were extremely weak due to the very small size of the crystal and hence the value of R is high. Refinement using reflections with $I > 3\sigma(I)$ led to a better value of R, but with an unacceptably low observation-to-parameter ratio. This refinement, however, yielded geometrical parameters very close to those of the determination reported here, but with greater e.s.d.'s. H atoms were placed in calculated positions and were not refined.

Data collection: *MSC/AFC Diffractometer Control Software* (Molecular Structure Corporation, 1988). Cell refinement: *MSC/AFC Diffractometer Control Software*. Data reduction: *TEXSAN PROCESS* (Molecular Structure Corporation, 1989). Program(s) used to solve structure: *MITHRIL* (Gilmore, 1994). Program(s) used to refine structure: *TEXSAN LS*. Molecular graphics: *ORTEP*II (Johnson, 1976). Software used to prepare material for publication: *TEXSAN FINISH*.

Lists of structure factors, anisotropic displacement parameters, H-atom coordinates and complete geometry have been deposited with the IUCr (Reference: VJ1013). Copies may be obtained through The Managing Editor, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

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Prolyl Endopeptidase Inhibitors. III. A Peptidyl α-Keto Benzothiazole Derivative

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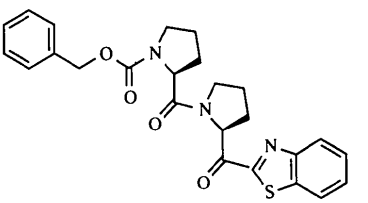
Abstract

In the title compound, benzyl 2-[2-(2-benzothiazoloyl)-pyrrolidinoyl]pyrrolidine-1-carboxylate, $C_{25}H_{25}N_3O_4S$, both the N-terminal urethane bond and the C-terminal amide bond are *trans*. The dipeptide inhibitor is semi-extended and shows coplanarity between the ketone carbonyl group and the benzothiazole ring, with the carbonyl O atom *cis* with respect to the ring S atom.

Comment

We are interested in the structures of peptidyl α-keto heterocyclic inhibitors of prolyl endopeptidase (PEP) (Tsutsumi *et al.*, 1994). We reported previously the structure of an α-keto thiazole inhibitor (Tsutsumi, Okonogi, Takeuchi & Kodama, 1995*b*). We report here the structure of an α-keto benzothiazole inhibitor, (I). This compound is as potent an inhibitor as the α-keto thiazole inhibitor.



(I)

The title dipeptide, with $\varphi_1 = -65\,(2)$, $\psi_1 = 150\,(1)$, $\varphi_2 = -65\,(1)$ and $\psi_2 = 155\,(1)°$ (IUPAC–IUB Commission on Biochemical Nomenclature, 1971), has a polyproline II conformation. Both the N-terminal urethane bond and the C-terminal amide bond are *trans* [O1—C12—N3—C11 173 (1) and C11—C10—N2—C9 176 (1)°]. In the two peptide inhibitors involving an N-terminal indolecarboxylic acid residue (Tsutsumi, Okonogi, Takeuchi & Kodama, 1994*a*,*b*), the corresponding bonds are *cis* and *trans*. Thus, the difference in the conformation appears to depend on the prolyl residue (Magaard, Sanchez, Bean & Moore, 1993) and not on the N-terminal moieties. The ketone carbonyl group and the benzothiazole ring are coplanar [O2—C8—C7—S1 −1 (2) and N1—C7—C8—O2 −180 (1)°]. The carbonyl O atom of the ketone group is *cis* with respect to the ring S atom.

The conformational relationship between the carbonyl O atom O2 and atom N1 of the heterocycle is *trans*, as in the α-keto thiazole inhibitor (Tsutsumi, Okonogi, Takeuchi & Kodama, 1994*b*). We believe that this conformation of peptidyl α-keto heterocyclic inhibitors is necessary in order to stabilize the hemiketal adduct of the active site.

A second aim of our work is to produce a drug with improved duration of *in vivo* action. The title compound and the α-keto thiazole derivative were orally

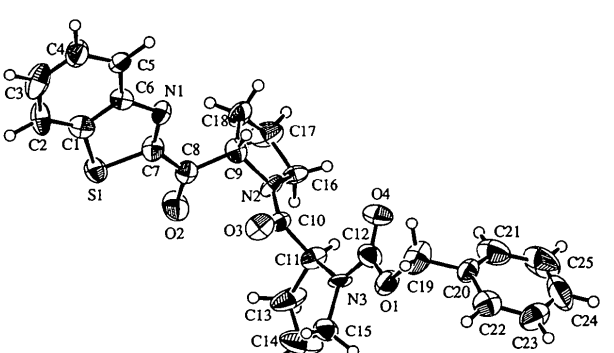


Fig. 1. The molecular structure of the title compound with the crystallographic numbering scheme (*ORTEP*II; Johnson, 1976). Displacement ellipsoids are shown at the 50% probability level and H atoms are drawn as spheres of arbitrary size.