Notes

Fibrin-Stabilizing Factor Inhibitors. 5. Primary Amines Related to Monotosylcadaverine[1]

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A series of primary amines related to monotosylcadaverine has been prepd and tested as fibrin-stabilizing factor (FSF) inhibitors. The results indicate that an efficient specific inhibitor of FSF should have the general structure, $ArX(CH_2)_5NH_2$, where X should be a strongly electron-attracting group conjugated with the aromatic moiety.

In previous papers of this series[2a-c] we have published a number of new compds which were tested as fibrin-stabilizing factor inhibitors (FSF inhibitors). From this work and from literature data[3,4] we have concluded that a specific FSF inhibitor should have the general formula **1**, where a pentyl chain, carrying a primary amino group at one end, is attached at its other end to an aromatic moiety (Ar) *via* a group X.

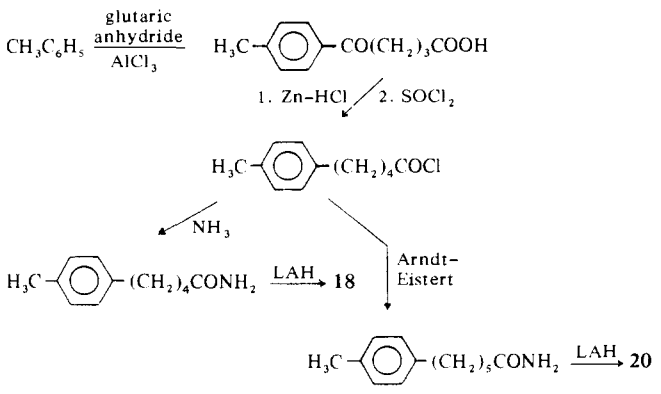
$$ArX(CH_2)_5NH_2$$
1

The primary amino group is essential for biologic activity[2c,3] and the pentyl chain seems to be the optimal length for the intervening chain.[2b,c,3] In this work we have investigated the influence of the group X on the activity.

Lorand, *et al.*,[3] have discovered that monodansylcadaverine and monotosylcadaverine (**2**) were potent inhibitors of FSF. Substance **2** was chosen as a base for our investigations. A series of compds was thus prepd where the SO_2NH moiety of **2** was replaced by other functional groups while the rest of the molecule was left intact.

Chemistry. The syntheses of the compds were generally accomplished in a straightforward manner as described in the Experimental Section. Compds **18** and **20** were obtained as indicated in Scheme I. The prepd compds are presented in Table I.

Scheme I



Bioassays. The biological assays were carried out as previously described[2a] and the inhibitory activity of each compd is expressed in relation to that of monodansylcadaverine (activity of monodansylcadaverine = 100).

Results and Discussion

The activities of the compds are collected in Table I. These data show that the sulfonamides **2**, **4**, and **5** have the highest activity of the compds tested. It is also apparent that among the compds with other acyl groups, those with a carbonyl function directly conjugated with the aromatic ring have the best activity (*cf.* **7** and **9**; **12** and **13**). The compds without the acyl group (**15**, **17**, **18**, **20**) generally have low activity.

It is of interest to note that the combination of a sulfonamide with an aromatic moiety is necessary for high FSF-inhibitory activity. This was evidenced by the observation that the compd $H_2NSO_2(CH_2)_5NH_2$, which has no aromatic function, has an activity of less than 1% of that of monodansylcadaverine. As the SO_2NH group is strongly electron attracting, the aromatic nucleus of **2**, **4**, and **5** is expected to have an electron density that is low in comparison to that of the other test compds. An electron-deficient aromatic nucleus in combination with the pentylamine as indicated in the general formula **1** thus appears to be a necessary structural requirement for high FSF-inhibitory activity.

The results of our studies obtained so far suggest that a specific FSF inhibitor should be attached to the enzyme near the active center *via* an electron-deficient aromatic nucleus. When the inhibitor is thus aligned in position, the side chain carrying the nucleophilic amino group must have a certain optimal length to permit the amino group to efficiently attack the carbonyl group of the thiol ester of the acyl-enzyme intermediate.[4] This reaction will form an amide bond between a γ-carboxyl group of a Glu residue of fibrin and the NH_2 group of the inhibitor, thus inhibiting the fibrin cross-linking.

Experimental Section

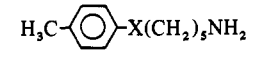
General Comments. Melting points were detd with calibrated Anschütz thermometers in an electrically heated metal block. All cryst compds were characterized by elemental analyses (C, H, N), which were within ±0.4% of the theoretical value, and ir spectra, which were run for identification purposes on a Perkin-Elmer 237 spectrophotometer.

***N*-(5-Aminopentyl)-*p*-toluenesulfonamide (monotosylcadaverine) (2)** was prepd from TsCl and cadaverine by the procedure described for monodansylcadaverine;[2c] yield 52%, mp for the hydrochloride 123–124° (from EtOH–Et_2O); lit.[3] mp 123.5–124.5°.

***N*-(4-Cyanobutyl)-*N*-methyl-*p*-toluenesulfonamide (3).** TsCl (0.50 g, 2.6 mmoles) in $CHCl_3$ (45 ml) was added to a soln of 5-methylaminovaleronitrile[5] (0.29 g, 2.6 mmoles) and Et_3N (1.0 g, 9.9 mmoles) in $CHCl_3$ (5 ml). The mixt was stirred at room temp for 15 hr and was then washed with 5% aqueous $NaHCO_3$ (2 × 50 ml) and with H_2O (2 × 50 ml), dried (Na_2SO_4), and evapd *in vacuo*. This yielded 0.65 g (94%) of a yellow oil which was used in the next step without further purification.

***N*-(5-Aminopentyl)-*N*-methyl-*p*-toluenesulfonamide (4).** Compd **3** (0.50 g, 1.9 mmoles) in EtOH (25 ml) and concd HCl (0.5 ml) was hydrogenated over PtO_2 at room temp at an initial pressure of 3 kg/cm² until the theoretical amt of H_2 had been consumed. After filtration and evapn of the filtrate, anhyd Et_2O (100 ml) and EtOH

Table I. Physical Data and Biological Properties of the Compounds Studied



Compd No.	X	Yield, %	Mp, °C	Formula	FSF-inhibitory activity[a]
2	SO_2NH	62	123-124	$C_{12}H_{20}N_2O_2S \cdot HCl$	28
4	SO_2NCH_3	60	106-107	$C_{13}H_{22}N_2O_2S \cdot HCl$	29
5	$NHSO_2$	34	147-149	$C_{12}H_{20}N_2O_2S \cdot HCl$	20
7	CONH	85	192-193	$C_{13}H_{20}N_2O \cdot HCl$	5
9	NHCO	74	180-182	$C_{13}H_{20}N_2O \cdot HCl$	2
11	NHCONH	83	182-183	$C_{13}H_{21}N_3O \cdot HCl$	5
12	CO_2	18	118-120	$C_{13}H_{19}NO_2 \cdot HCl$	8
13	OCO	81	132-134	$C_{13}H_{19}NO_2 \cdot HCl$	3
15	NH	75	211-213	$C_{12}H_{20}N_2 \cdot 2HCl$	2
17	$NHCH_2$	70	214-215	$C_{13}H_{22}N_2 \cdot 2HCl$	2
18		62	157-159	$C_{12}H_{19}N \cdot HCl$	<1
20	CH_2	45	148-150	$C_{13}H_{21}N \cdot HCl$	<1

[a] In per cent of that of monodansylcadaverine, determined as previously described.[2a]

(5 ml) were added. The pptd hydrochloride was collected and recryst from EtOH-Et_2O; yield 0.35 g (60%), mp 106-107°. *Anal.* ($C_{13}H_{22}N_2O_2S \cdot HCl$) C, H, N.

5-(*p*-Tolylsulfamoyl)pentylamine (5) was prepared as previously described.[2b]

***N*-(4-Cyanobutyl)-*p*-methylbenzamide (6).** To a soln of *p*-toluoyl chloride (0.47 g, 3.0 mmoles) in anhyd Et_2O (50 ml) was added 5-aminovaleronitrile[5] (0.59 g, 6.0 mmoles). The mixt was stirred at room temp for 1 hr and the pptd product was filtered off and washed with cold H_2O; yield 0.60 g (92%), mp 66-67° (from EtOH-H_2O). *Anal.* ($C_{13}H_{16}N_2O$) C, H, N.

***N*-(5-Aminopentyl)-*p*-methylbenzamide (7).** Compd **6** (0.50 g, 2.3 mmoles) was hydrogenated over PtO_2 as described for **4**. This yielded 0.50 g (85%) of the hydrochloride, mp 192-193° (from Me_2CO-EtOH). *Anal.* ($C_{13}H_{20}N_2O \cdot HCl$) C, H, N.

6-Benzyloxycarbonylaminohexanoyl-*p*-toluidide (8) was prepd from *N*-benzyloxycarbonyl-6-aminohexanoic acid[6] and *p*-toluidine by the mixed anhydride method as previously described;[2a] yield 72%, mp 117-118° (from toluene). *Anal.* ($C_{21}H_{26}N_2O_3$) C, H, N.

6-Aminohexanoyl-*p*-toluidide (9). Compd **8** was hydrogenolyzed over Pd/C as previously described,[2a] affording a yellow oil which was converted to the hydrochloride; yield 74%, mp 180-182° (from EtOH-Et_2O). *Anal.* ($C_{13}H_{20}N_2O \cdot HCl$) C, H, N.

***N*-(4-Cyanobutyl)-*N'*-*p*-tolylurea (10).** To a soln of *p*-tolyl isocyanate (0.68 g, 5.1 mmoles) in anhyd Et_2O (50 ml) was slowly added 5-aminovaleronitrile[5] (0.50 g, 5.1 mmoles). The mixt was stirred for 1 hr at room temp and the product collected; yield 0.90 g (76%), mp 136-138° (from toluene). *Anal.* ($C_{13}H_{17}N_3O$) C, H, N.

***N*-(5-Aminopentyl)-*N'*-*p*-tolylurea (11).** Compd **10** (0.46 g, 2.0 mmoles) was hydrogenated over PtO_2 as described for **4**, affording 0.45 g (83%) of the hydrochloride, mp 182-183° (from Me_2CO-EtOH). *Anal.* ($C_{13}H_{21}N_3O \cdot HCl$) C, H, N.

5-Aminopentyl *p*-Toluic Acid Ester (12). 5-Aminopentanol (2.1 g, 20 mmoles) was added to a soln of *p*-toluoyl chloride (3.1 g, 20 mmoles) and AcOH (1.2 g, 20 mmoles) in dry C_6H_6 (100 ml) and the mixt refluxed overnight. After cooling, Et_2O (100 ml) was added and the pptd product filtered off, suspended in 50 ml of 1 *M* Na_2CO_3, and extracted with $CHCl_3$ (3 × 50 ml). The organic ext was dried (Na_2SO_4) and evapd *in vacuo*, affording an oil which was dissolved in EtOH-Et_2O and pptd as hydrochloride; yield 0.90 g (18%), mp 118-120° (from EtOH-Et_2O). *Anal.* ($C_{13}H_{19}NO_2 \cdot HCl$) C, H, N.

***p*-Tolyl 6-Aminohexanoic Acid Ester (13).** A mixt of 6-aminohexanoyl chloride·HCl[7] (4.5 g, 2.4 mmoles) and *p*-cresol (2.6 g, 2.4 mmoles) in dry C_6H_6 (75 ml) was stirred overnight at room temp. Anhyd Et_2O (100 ml) was then added and the pptd product filtered off and washed with Et_2O; yield 5.0 g (81%), mp 132-134° (from C_6H_6). *Anal.* ($C_{13}H_{19}NO_2 \cdot HCl$) C, H, N.

***N*-(4-Cyanobutyl)-*p*-toluidine (14).** A soln of 5-chlorovaleronitrile (11.7 g, 100 mmoles) and *p*-toluidine (21.4 g, 200 mmoles) in *p*-xylene (250 ml) was refluxed for 24 hr. After cooling, the filtered soln was washed with 5% aqueous $NaHCO_3$ (2 × 150 ml) and with H_2O (2 × 150 ml), dried (Na_2SO_4), and evapd *in vacuo*. The residue was distd *in vacuo*, yielding 12.0 g (64%) of product, bp 156-158° (0.5 mm), mp 53-55° (from ligroin-EtOH). *Anal.* ($C_{12}H_{16}N_2$) C, H, N.

***N*-(5-Aminopentyl)-*p*-toluidine (15).** The nitrile **14** (2.0 g, 10.6 mmoles) was refluxed with LAH (0.42 g, 11 mmoles) in anhyd Et_2O (150 ml) for 6 hr and then stirred overnight at room temp. Aqueous 5 *M* NaOH (30 ml) was then added, and the Et_2O layer sepd. The alk phase was extd with Et_2O (3 × 100 ml) and the entire Et_2O extracts were dried (Na_2SO_4) and evapd. The residue was distd *in vacuo*, yielding a yellow oil, bp 155-160° (1.5 mm); mp of the dihydrochloride 211-213° (from EtOH-E_2O), yield 2.1 g (75%). *Anal.* ($C_{12}H_{20}N_2 \cdot 2HCl$) C, H, N.

***N*-(5-Cyanopentyl)-*p*-toluidine (16)** was prepd from 6-bromocapronitrile (4.5 g, 25.6 mmoles) and *p*-toluidine (5.5 g, 51 mmoles) as described for **14**; yield 3.1 g (60%), mp 82-83° (from ligroin-EtOH). *Anal.* ($C_{13}H_{18}N_2$) C, H, N.

***N*-(6-Aminohexyl)-*p*-toluidine (17)** was prepd as described for **15**; yield 70%, mp of the dihydrochloride 214-215° (from EtOH-Et_2O). *Anal.* ($C_{13}H_{22}N_2 \cdot 2HCl$) C, H, N.

5-*p*-Tolylpentylamine (18). 5-*p*-Tolylpentanamide[8] (0.50 g, 5 mmoles) was reduced with LAH (0.19 g, 5 mmoles) in anhyd Et_2O (150 ml) for 20 hr. The mixt was worked up as usual yielding a yellow oil which was pptd as the hydrochloride; mp 157-159° (from Me_2CO), yield 0.35 g (62%). *Anal.* ($C_{12}H_{19}N \cdot HCl$) C, H, N.

6-*p*-Tolylhexanamide (19). To an ice-cold soln of CH_2N_2 (1.25 g, 30 mmoles) in Et_2O (60 ml) was slowly added a soln of 5-(*p*-tolyl)-pentanoyl chloride[9] (2.0 g, 9.5 mmoles) in Et_2O (15 ml) over a period of 10 min. The mixt was stirred at room temp overnight and the Et_2O was distd off. Dioxane (25 ml), concd NH_3 (15 ml), and 10% aqueous $AgNO_3$ (3 ml) were added to the residual yellow oil and the mixt was refluxed for 2 hr. After cooling, ice (100 g) was added and the pptd amide collected; yield 1.15 g (59%), mp 119.5-120.5° (lit.[10] mp 118.5°).

6-(*p*-Tolyl)hexylamine (20). This compd was prepd analogously to **18**; yield 45%, mp of the hydrochloride 148-150° (from Me_2CO). *Anal.* ($C_{13}H_{21}N \cdot HCl$) C, H, N.

5-Aminopentanesulfonamide was prepd as previously described.[11]

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