Central Nervous System Activities of Thiolactams and Lactams. Structure–Activity Correlations†

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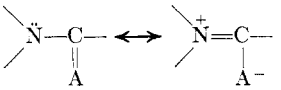
The CNS activities of five thiolactams were studied for the first time. These compounds caused clonic and tonic convulsions in mice a few sec after ip injection. The most potent compd examined is 2-azacyclooctanethione which has LD_{50} 23 mg/kg. The variation of the acute lethal toxicity (log $1/LD_{50}$) of series of thiolactams was found to be mainly due to the variation in the lipophilic character as measured by the octanol–H_2O partition coefficient or tlc (log P or $-R_M$). The dipole moment (μ) term appears to play a significant role in determining the convulsant activity and the toxicity of thiolactams, lactams, and γ-thiobutyrolactone.

Many organic compounds containing the amide or thioamide type of resonating structures



(where A = O,S) have been found to possess CNS activities.[1,2] Although thiolactams have been known for more than a decade, no systematic study of their biological activities has been reported thus far.

As an extension of our study on the CNS activities of lactams,[3] cyclic ureas, and thioureas,[4] the authors wish to report the more potent CNS stimulatory activity and the structure–activity correlation of a series of thiolactams and lactams.

Experimental Section

Materials.—The thiolactams were synthesized in our laboratory by refluxing the corresponding lactams with thioacetic acid.[5] The mp of the recrystd products agreed with the lit. values. The elemental anal. were performed by C. F. Geiger, Ontario, Calif.; the experimental values of C, H, N, and S were all within ±0.4% of the calcd values.

Acute Lethal Toxicity (LD_{50}).—Groups of 6 Swiss–Webster albino mice, 3 males and 3 females weighing 18–28 g, were injected ip with a series of doses. Signs of CNS activity were observed continuously for 2 hr and then at regular intervals for 2 days. All drugs were administered in aq soln in a vol of 1 ml or less unless otherwise stated. Control animals received saline injection. The LD_{50} was estimated for each drug according to the method of Miller and Tainter.[6]

Measurement of the Partition Coefficient.—The 1-octanol–H_2O partition coefficients (P) were measured using a Perkin-Elmer Model-124 double beam grating spectrophotometer. The octanol was satd with H_2O and *vice versa*, so no corrections for vol change after equil were necessary. The analytical wavelength ranged from 260 to 272 mμ (see Table I).

Tlc.—Two different solvent systems were used in this study, *i.e.*, dioxane and BuOH–AcOH–H_2O (4:2:1). Baker-flex sheets precoated with silica gel-1B were used to measure the R_f values of the thiolactams. These R_f values were converted to the R_M values according to the formula $R_M = \log (1 - R_f)/R_f$ or the conversion table.[7]

Regression Analysis.—The method of least squares was used in correlating the acute lethal toxicity (log $1/LD_{50}$) and the physicochemical constants using an IBM 360/65 computer.

Effect of 2-Azacyclooctanethione on the Motor Activity of Mice.—2-Azacyclooctanethione (3 mg/kg) was injected into each mouse, and the animal was placed in an Electronic Motility Meter Fc 40 (Mortron Produkter, Stockholm, Sweden) enclosed with a circular wall with 20 cm diameter. The cumulative reading over a 30-min period after injection was compared with that of a control animal receiving normal saline injection. The results of 6 test mice (3 males and 3 females) as compared with the results of 6 mice in the control group are summarized in Table III.

Effect of 2-Azacyclooctanethione on the Pentobarbital Sleeping Time in Mice.—A group of 6 mice (3 males and 3 females) were given 15 mg/kg of the drug 10 min after the injection of sodium pentobarbital (60 mg/kg), and the sleeping times were compared with those of the animals receiving sodium pentobarbital alone. The results are shown in Table III.

Results and Discussion

The signs observed and the estimated LD_{50} values as well as the physicochemical constants used in the correlation are assembled in Table I. The equations correlating the toxicity and the physicochemical parameters are summarized in Table II.

All the thiolactams studied caused clonic and tonic convulsions within a few sec of ip injection. The 5-membered (n = 3) thiolactam caused mild sedation at lower doses and convulsions at higher doses. Since the dual activities were observed for the 7-membered lactam, it appeared that this was probably not due to conformational difference. From ir study it has been shown[8] that the cis–trans transition occurred for the ring size of 9. The most potent compd studied is 2-azacyclooctanthione (LD_{50} = 0.16 mmole/kg), which is more toxic than pentylenetetrazole (LD_{50} = 0.67 mmole/kg) and nikethamide (LD_{50} = 0.98 mmole/kg), but less toxic than strychnine LD_{50} = 0.003 mmole/kg) or picrotoxin (LD_{50} = 0.012 mmole/kg) in mice by the same route of administration.[9]

From eq 1, 4, and 5 one can see that the variation in the toxicity of the thiolactams is mainly due to the change in the lipohydrophilic character as measured by log P or $(-R_M)$. More than 85% ($\gamma^2 > 0.85$) of the variance in the data can be accounted for by eq 1, 4, or 5. Addition of the dipole moment (μ) term in eq 2 did not significantly improve the correlation. Combination of the $-R_M$ and μ terms did not further

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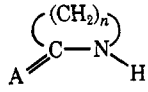
Table I

Acute Lethal Toxicity and Physicochemical Constants of Thiolactams and Lactams



n	Mp, °C cor	Conformation[a]	Observation	LD_{50}, mg/kg	log 1/C, moles/kg	μ (D)	λ_{max}, mμ	log P ± S.D.	R_M BuOH-AcOH-H_2O	R_M Dioxane
			A = S							
3	112.5–114 (112–113)	Cis (strain)	Mild sedat at 500 mg/kg; conv and saliv at 1000 mg/kg	525	2.28	4.52[b] (5.07)[c]	260	−0.05 ± 0.03	0.503	0.676
4	94.5–95.5[d] (95–96)[d]	Cis (half-chair, strainless envelop)	Convulsions	175	2.82	4.83[b] (5.15)[c]	267	0.13 ± 0.02	0.555	0.760
5	104–105.5 (105–105.5)[d]	Cis (envelop; boat)	Convulsions	65	3.30	4.88[b] (4.83)[e]	270	0.75 ± 0.02	0.743	0.822
6	81.5 (82–83)[d]	Cis (puckered ring; boat)	Convulsions, salivation, urination	23	3.80	4.86[b] (4.81)[e]	272	1.00 ± 0.02	0.747	0.841
7	87.5 (87.5–88)[d]	Cis (skew)	Convulsions, salivation, urination	30[f]	3.72	4.85[b] (4.80)[e]	272	1.44 ± 0.01	0.814	0.866
			A = O							
5		Cis (envelop; boat)	Slight sedat at 300 mg/kg, convulsions at 500 mg/kg	650[g]	2.24[g]	3.88[h]		−0.19[g]		
6		Cis (puckered ring boat)	Convulsions	270[g]	2.67[g]	3.86[h]		0.24[g]		
7		Cis (skew)	Convulsions	185[g]	2.88[g]	3.85[h]		0.67[g]		
11		Trans	Trembling, side position	625[g,i]	1.72[g]	3.64[h]		1.72[g]		
			γ-Thiobutyrolactone							
			Convulsions	199[g]	2.71[g]	3.83[j]		0.60[g]		

[a] From H. E. Hallam and C. M. Jones, *J. Chem. Soc. A*, 1033 (1969). [b] Estimated by adding 1 Debye unit to the dipole moment of the corresponding lactam measured in C_6H_6 at 25°. [c] From C. M. Lee and W. D. Kumler, *J. Org. Chem.*, **27**, 2052 (1962), measured in dioxane at 30°. [d] From R. Huisgen and J. Witte, German Patent 1,024,508 (1958); *Chem. Abstr.*, **54**, 19453c (1960). [e] Estimated value based on the dipole moment of the 7-membered thiolactam (n = 5) and the differences of the dipole moments of 7- to 9-membered lactams. [f] Propylene glycol (1%) was added to prevent precipitation of the drug, the vehicle alone did not show any CNS activity. [g] From ref 3. [h] From A. L. McClellan, "Tables of Experimental Dipole Moments," W. H. Freeman and Co., San Francisco and London, 1963, measured in benzene at 25°. [i] Suspended in 5% gum acacia. [j] From I. Wallmark, M. H. Krackov, S. H. Chu, and H. G. Mantner, *J. Amer. Chem. Soc.*, **92**, 4447 (1970), measured in C_6H_6 at 25°.

Table II

Equations Correlating Acute Lethal Toxicity and Physicochemical Constants of the Thiolactams and Lactams

Compds	Equation	n[a]	r[b]	s[c]	log P_0 (± 95% c.i.)	Eq. no.
Thiolactams	$\log 1/C = 0.940 \log P + 2.583$	5	0.929	0.272		1
	$\log 1/C = 0.695 \log P + 1.582\mu - 4.941$	5	0.972	0.212		2
	$\log 1/C = -0.633(\log P)^2 + 1.764 \log P + 2.516$	5	0.963	0.244	1.39 (± ∞)	3
	$\log 1/C = 7.571(-R_M)_{diox}$[d] $- 2.850$	5	0.955	0.193		4
	$\log 1/C = 4.460(-R_M)_{BAW} - 0.185$	5	0.944	0.243		5
	$\log P = 7.253(-R_M)_{diox} - 5.127$	5	0.959	0.203		6
	$\log P = 4.053(-R_M)_{BAW} - 2.033$	5	0.968	0.180		7
	$(-R_M)_{BAW} = 1.604(-R_M)_{diox} - 0.606$	5	0.966	0.041		8
Lactams + γ-thiobutyrolactone	$\log 1/C = 0.083 \log P + 2.550$	5	0.243	0.272		9
	$\log 1/C = 0.969 \log P + 6.834\mu - 24.028$	5	0.968	0.086		10
	$\log 1/C = -0.506(\log P)^2 + 0.903 \log P + 2.441$	5	0.970	0.083	0.89 (0.59–1.46)	11
Thiolactams	$\log 1/C = 0.474 \log P + 2.596$	10	0.547	0.488		12
Lactams and γ-thiobutyrolactone	$\log 1/C = 0.462 \log P + 0.685\mu - 0.342$	10	0.856	0.342		13
	$\log 1/C = -0.802(\log P)^2 + 1.660 \log P + 2.458$	10	0.760	0.405	1.04 (± ∞)	14
	$\log 1/C = -0.589(\log P)^2 + 1.336 \log P + 0.584\mu - 0.012$	10	0.935	0.240	1.13 (0.85–8.00)	15

[a] The number of data points used in the regression analysis. [b] The correlation coefficient. [c] The standard deviation from regression. [d] Diox, dioxane; BAW, BuOH-AcOH-H_2O.

improve the correlation either. The $(\log P)^2$ term in eq 3 is not statistically significant; nevertheless, this equation reflects the fact that the optimum lipophilic character for maximum toxicity (log P_0, obtained by setting d (log 1/C)/d log P = 0) is around 1.4.

When the additivity of the π constant (π_x = log $P_{\text{parent mol}-x}$ − log $P_{\text{parent mol}}$ was examined as the ring size increased from 5 to 9 (n = 3–7), it was found that the π_{CH_2} ranged from 0.18 to 0.62 instead of staying around 0.42 ($\pi_{CH_2} = {}^1/_6\pi_{cyclohexene} = 0.42$).[10] Since the standard deviation of the experimental log P values is of the order of 0.01 to 0.03, this variation of π_{CH_2} values probably is not due to experimental error. In order to verify this, the log P, the $-R_M$, and the μ values are plotted *vs.* n (Figure 1).

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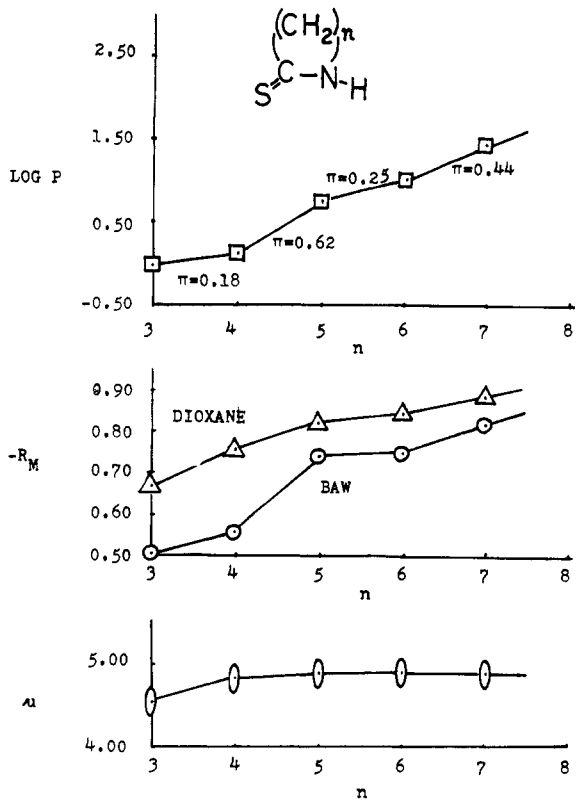

Figure 1.—The dependence of the physicochemical constants of thiolactams on the ring size.

From this figure one can see that the change in log P more or less parallels the change of $-R_M$. This is also indicated by the high correlation coefficients of eq 6 and 7. The surprisingly low log P of the 6-membered ring ($n = 4$) may be attributed to its relatively high dipole moment as compared with that of the 5-membered ring, resulting in higher water solubility. For the 7-membered and larger rings the dipole moments stay pretty constant and the π_{CH_2} is closer to 0.4. This finding points to the importance of the electronic effect on the additive-constitutive nature of the π constant. One can also see that, at least in a number of cases, the $-R_M$ values give practically the same correlation as log P,[11] and the R_M values obtained from different solvent systems correlate quite well (eq 8).

For the 4 lactams and γ-thiobutyrolactone, the dipole moment term in eq 10 is highly significant ($F_{1,2} = 27$), so is the $(\log P)^2$ term in the parabolic eq 11 ($F_{1,2} = 30$). All the dipole moments of the lactams were experimentally determined;[12] unfortunately, only 3 experimental dipole moments of the thiolactams were available in the literature.[13] The reported dipole moment of the thiolactams were measured in dioxane and those of the lactams were measured in C_6H_6. It is known that dioxane solutions usually give higher dipole moments than C_6H_6 solutions;[14] in order to avoid this additional source of error we elected to use the estimated dipole moment of the thiolactams by adding 1 Debye unit to the μ of the corresponding lactam measured in C_6H_6. These 5 estimated values gave better correlation than the 3 experimental values plus 2 estimated values.

When the thiolactams, lactams, and γ-thiobutyrolactone are pooled together, eq 12–15 are obtained. The $(\log P)^2$ and μ terms in eq 15 are significant at 95 percentile level ($F_{1,6} = 6.6$; 13 resp). More than 87% ($\gamma^2 > 0.87$) of the variance in the data can be "explained" by eq 15. It is interesting to note that the log P_0 value of 1.13 obtained from eq 15 is very close to those that have been found for a series of cyclic urea and thiourea derivatives (log $P_0 = 1.4$–1.5).[4] This suggests that the site of action and the intermolecular forces involved in drug–receptor interactions are probably the same for all the aforementioned compounds. The sign associated with the μ term in eq 2, 10, 13, and 15 is positive, indicating that the higher the dipole moment the higher will be the convulsant activity and the acute lethal toxicity. This is in accord with what has been found for the acute lethal toxicity of a series of cyclic urea and thiourea derivatives,[4] and in contrast with what has been found for the anticonvulsant activity of a series of imides, barbiturates, and hydantoins,[15] where the dependence of the anticonvulsant activity on the μ term is negative.

From Table III it can be seen that even though the motor activity was significantly increased by 3 mg/kg of 2-azacyclooctanethione, the pentobarbital-induced sleeping time was increased rather than decreased by 15 mg/kg of the thiolactam. This may possibly be due to competitive protein binding, thus releasing more of the pentobarbital for CNS depression. This finding ruled out the possibility of using the thiolactams as possible analeptic agents in barbiturate poisoning. One possible application of the potent thiolactams may be the use as rodenticides. In preliminary testing when some cheese was incorporated with 2-azacyclooctanthione and given to 6 mice without giving the regular food, 3 of the animals died within 24 hr and all of them died in 2 days.

TABLE III

EFFECTS OF 2-AZACYCLOOCTANETHIONE ON THE MOTOR ACTIVITY AND THE SODIUM PENTOBARBITAL SLEEPING TIME IN MICE

Drug(s) given	Motor activity ± S.D.	Sleeping time,[a] min ± S.D.	% increase	t	P
Saline (control)	849 ± 67				
2-Azacyclooctanethione (3 mg/kg)	1146 ± 272		+35	2.37	<0.05
Sodium pentobarbital (60 mg/kg)		52 ± 26			
Sodium pentobarbital (60 mg/kg) + 2-azacyclooctanethione (15 mg/kg)		87 ± 19	+65	2.35	<0.05

[a] The sleeping time was arbitrarily defined as the time interval between the loss and the spontaneous recovery of the righting reflex.

Acknowledgments.—The authors wish to thank the staff of the Computer Science Laboratory of this University for the data processing.

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