the free base was eluted with ethyl acetate. Reconversion of the free base to the hydrochloride salt with ethereal hydrogen chloride and recrystallization from acetone–ether provided pale-yellow crystals, mp 108–110° dec.

Anal.—Calc. for $C_{21}H_{32}Cl_2N_2O_4$: C, 56.37; H, 7.21; N, 6.26. Found: C, 56.53; H, 7.20; N, 6.11.

Pharmacology—The blood pressure lowering activities of the compounds studied were compared to papaverine hydrochloride, the standard, in three male cats weighing an average of 3 kg. The animals were anesthetized with pentobarbital sodium (30 mg/kg), with additional small doses being given as needed. The blood pressure was monitored continuously by a force transducer[2] connected to a polyethylene catheter placed in the femoral artery and coupled[3] to a recording polygraph[4] at a chart speed of 1 cm/min. The average blood pressures for the cats at the beginning and end of the experiments were 146 and 132 mm/Hg, respectively.

The compounds tested were simply dissolved in normal saline if they were in a salt form. If in the free base form, they were solubilized in the least amount of 0.02 *N* hydrochloric acid and made up to the normal saline concentration with appropriate amounts of sodium chloride and water. The compounds tested were injected as a bolus *via* a femoral venous catheter in doses ranging from 0.5 to 3 mg/kg, followed by a 3-ml saline flush. The results reported (Table I) for each dose are the results of three or more readings. The blood pressure was allowed to return to preinjection levels before additional injections were given.

The protocol for the administration of the several agents being tested was to administer them according to the Latin-square method, whereby each new animal preparation was tested with a different order of the agents. In addition, administration of papaverine hydrochloride at the beginning, during the course of, and at the end of the experiment indicated that the blood pressure was still normally responsive in each cat throughout the experiment. The drop in blood pressure by an average dose of 1.67 mg/kg of papaverine hydrochloride was 60 mm/Hg, and this value was assigned as a 100% drop in the blood pressure because increasing the dose failed to elicit a further drop. All compounds were rated as a percentage against the standard on this basis.

[2] Beckman 215071.
[3] Beckman 9853 coupler.
[4] Beckman R-411 dynograph.

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Cyanogen Condensations as a Route to 3-Amino-2-imino-1,3-benzothiazin-4-ones with CNS Depressant Potential

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Abstract □ Seven new 3-amino-2-imino-1,3-benzothiazin-4-ones, prepared from the condensation of cyanogen and 2-mercaptobenzhydrazides, were evaluated in a neuropharmacological mouse profile. CNS depression was observed in several members of the class.

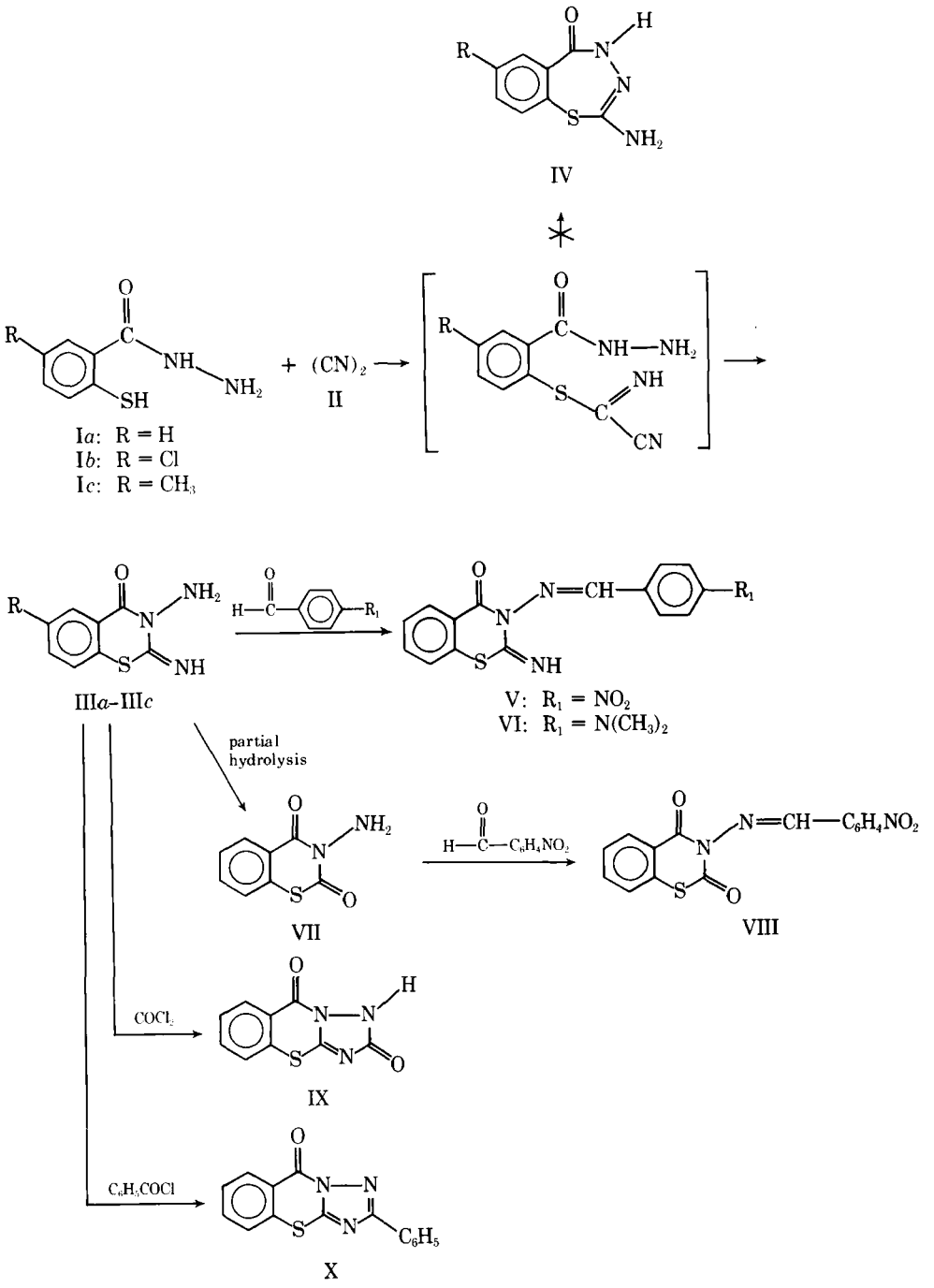
Keyphrases □ 3-Amino-2-imino-1,3-benzothiazin-4-ones—synthesized and screened as CNS agents □ CNS agents, potential—synthesis and screening of seven 3-amino-2-imino-1,3-benzothiazin-4-ones □ 1,3-Benzothiazin-4-ones, 3-amino-2-imino—synthesized and screened as CNS agents

Recent interest in the synthesis and pharmacological properties of certain 1,3-benzothiazin-4-ones, particularly as central nervous system (CNS) agents with marked mydriatic activity (1–5), prompted this report on a new synthesis of some uniquely substituted analogs of this class and their biological activities. 2-Mercaptobenzhydrazides (I) and cyanogen (II) were found to undergo facile condensation, in 50–74% yield, to 3-amino-2-imino-3,4-dihydro-2*H*-1,3-benzothiazin-4-ones (III), a reaction that represents cyanogen acting as a one-carbon insertion unit to close a six-membered ring (Scheme I).

Previous examples are known in which cyanogen in heterocyclic syntheses completes a cycle by insertion of a single carbon (6), two carbons (7), or one carbon and a nitrogen (8). An earlier synthetic study with 2-mercaptobenzhydrazide reported its condensation with formaldehyde to form a seven-membered ring, but no biological testing data were provided on the resulting benzo-1,3,4-thiadiazepinone (9).

The benzothiazinone products, III*a*–III*c*, could be structurally distinguished from the equally plausible, and isomeric, seven-membered alternatives, IV*a*–IV*c*, by characteristic chemical reactions. Hydrolysis generated Compound VII, which possessed a free amino group as evidenced by formation of an anil, VIII. Furthermore, condensation of III*a* with phosgene and/or benzoyl chloride produced *s*-triazolo systems, which would have been impossible from the alternative structure, IV. In addition, the pendant amino on III*a* underwent facile anil formation with both *p*-nitro- and *p*-dimethylaminobenzaldehydes.

From a practical viewpoint, solutions of up to 5 *M* cyanogen in anhydrous tetrahydrofuran, stable to



Scheme I

prolonged storage, are readily prepared by dissolving measured volumes of the condensed cyanogen into the solvent. The hydrogen cyanide gas evolved during the condensations of cyanogen with *Ia–Ic* is collected in a caustic trap and the heterocyclic products, IIIa–IIIc, precipitate directly in the tetrahydrofuran solution, not as salts but as their free base forms.

RESULTS[1]

Compounds IIIa, IIIb, IIIc, IIIa-HCl, V, VI, and VII were administered intraperitoneally in solution or as a suspension in water–methylcellulose to groups of four mice. Physiological signs were observed and evaluated according to the Irwin method (10). Compound IIIa and its hydrochloride salt, IIIa-HCl, gave virtually identical profiles with the exception of the rapidity of onset of the physiological effects. Both agents effected tonic convulsions and death at any dose above 30 mg/kg with an estimated LD_{50} of 18 mg/kg. Presumably because of accelerated absorption from the peritoneal cavity, IIIa-HCl caused death in less than 3 min post-dosing with 300 mg/kg while IIIa required 10 min. All other signs—hypothermia (4° at 100 mg/kg), salivation, body drop, Staub tail phenomenon, and loss of righting reflexes and of spontaneous motor activity—which were common to both agents, appeared more rapidly in IIIa-HCl.

The methyl analog IIIc gave a similar profile to IIIa but was considerably less toxic (LD_{50} of 178 mg/kg) and less active. At 100 mg/kg, IIIc functioned as a mild CNS depressant sedative with marked reduction in spontaneous motor activity, loss of righting reflexes, and hypothermia (4°). The chloro compound IIIb was

[1] Evaluations were carried out at Pharmakon Laboratories, Inc., Scranton, Pa., under the supervision of Dr. Richard Matthews.

nontoxic and inactive at doses up to 300 mg/kg. Therefore, the nature of the substituent at C-6 appears to influence activity markedly.

The 2-imino moiety apparently is involved in the toxic and depressant effects since there was no activity or toxicity with the hydrolyzed carbonyl counterpart, VII, at the highest dose tested, *i.e.*, 1000 mg/kg. Conversion of the 3-aminobenzylidene to a 4-dimethylaminobenzylidene led to a nontoxic compound, VI, which still evidenced mild catatonia and loss of motor activity at the maximum dose employed of 300 mg/kg. The 4-nitrobenzylidene compound, V, had an LD_{50} of 240 mg/kg; at 100 mg/kg, it too displayed mild CNS depressant effects including abnormal gait, lacrimination, hypothermia, and loss of motor activity. None of the compounds was judged sufficiently active to merit more extensive studies.

EXPERIMENTAL[2]

2-Mercaptobenzhydrazides (Ia–Ic)—The method of Katz *et al.* (9), involving thiolation of the diazonium salt of the corresponding anthranilic acid, esterification of the resulting 2-mercaptobenzoic acid, and its displacement with hydrazine, was employed in the syntheses of the 2-mercaptobenzhydrazides, Ia and Ib. The preparation and properties of Ic have not been reported previously, but by following the exact procedure for conversion of 5-chloroanthranilic acid to Ib, 5-methylanthranilic acid was converted in 26% yield to 2-mercapto-5-methylbenzhydrazide (Ic), mp 146–148°; IR (KBr): 3240 (br) (bonded N—H), 2560 (S—H), and 1610–1580 (C=O) cm^{-1}.

Anal.—Calc. for $C_8H_{10}N_2OS$: C, 52.73; H, 5.53; N, 15.37. Found: C, 52.58; H, 5.57; N, 15.36.

3-Amino-2-imino-3,4-dihydro-2*H*-1,3-benzothiazin-4-ones (IIIa–IIIc)—To a solution of 5.2 g (0.10 mole) of II, 4 drops of triethylamine, and 20 ml of anhydrous tetrahydrofuran at 0° was added dropwise a solution of 1–4 mmoles of the mercaptobenzhydrazide, Ia–Ic, in 65 ml of anhydrous tetrahydrofuran. Crystallization of the product commenced almost immediately, and the mixture was stirred at 5° for 4 hr after the completion of the addition. Dilution of the medium with 100 ml of benzene completed the precipitation of product, which was filtered and recrystallized from benzene. A 74% yield of IIIa, mp 141.5–142.5°, was obtained.

Anal.—Calc. for $C_8H_7N_3OS$: C, 49.73; H, 3.65. Found: C, 49.63; H, 3.54.

A 50% yield of IIIb, mp 144–145°, was obtained.

Anal.—Calc. for $C_8H_6ClN_3OS$: C, 42.20; H, 2.66. Found: C, 42.26; H, 2.62.

A 67% yield of IIIc, mp 154–155°, was obtained.

Anal.—Calc. for $C_9H_9N_3OS$: C, 52.16; H, 4.38. Found: C, 52.01; H, 4.35.

A hydrochloride salt of IIIa was prepared by introducing hydrogen chloride gas into a saturated solution of IIIa in benzene to precipitate IIIa-HCl, mp 206–209°; IR (KBr): 2600 broad (NH salt) cm^{-1}.

Anal.—Calc. for $C_8H_7N_3OS$·HCl: Cl, 15.44. Found: Cl, 15.15.

3-Arylideneimino-2-imino-3,4-dihydro-2*H*-1,3-benzothiazin-4-ones (V and VI)—A solution of 5.0 mmoles of IIIa and 5.0 mmoles of *p*-nitrobenzaldehyde in 50 ml of methanol containing 0.5 ml of acetic acid was refluxed for 2 hr, concentrated *in vacuo* to 20 ml, and filtered. The yellow solid (1.2 g, 76%) was recrystallized from dimethyl sulfoxide to yield the analytical sample of V, mp 307.5–308.5°.

Anal.—Calc. for $C_{15}H_{10}N_4O_3S$: N, 17.17. Found: N, 17.35.

In a similar fashion, 5.0 mmoles of IIIa and 5.0 mmoles of *p*-dimethylaminobenzaldehyde were reacted to yield VI (0.80 g, 48%), mp 264–265°.

Anal.—Calc. for $C_{17}H_{16}N_4OS$: N, 17.27. Found: N, 17.55.

3-Amino-3,4-dihydro-2*H*-1,3-benzothiazin-2,4-dione (VII)—A hydrolysis solution of 2 ml of acetic acid, 35 ml of ethanol, 10 ml of chloroacetaldehyde, and 10 ml of water was employed to dissolve 1.9 g (0.01 mole) of IIIa. The solution was then warmed to 45–55° and held at that temperature for 20 hr, during which time a buff-white solid precipitated. This solid, VII (1.07 g, 55%), was filtered and recrystallized from methanol, mp 199.0–199.5°; IR (KBr): 3310, 3245 (NH), and 1688 (C=O) cm^{-1}.

Anal.—Calc. for $C_8H_6N_2O_2S$: C, 49.48; H, 3.11; N, 14.42. Found: C, 49.85; H, 3.22; N, 14.80.

3-(4-Nitrobenzylideneimino)-3,4-dihydro-2*H*-1,3-benzothiazin-2,4-dione (VIII)—After refluxing equimolar (1.0 mmole) amounts of VII and *p*-nitrobenzaldehyde in 20 ml of methanol containing 5 drops of acetic acid for 1 hr, a 67% yield of VIII was obtained. Recrystallization from methanol gave the analytical sample, white microneedles, mp 220.5–221.5°; IR (KBr): no N—H bands, 1690 (C=O) cm^{-1}.

Anal.—Calc. for $C_{15}H_9N_3O_4S$: C, 55.04; H, 2.77; N, 12.84. Found: C, 54.87; H, 2.81; N, 12.79.

1,2-Dihydro-*s*-triazolo[5,1-*b*]benzo[1,3]-thiazin-2,9-dione (IX)—A well-stirred solution of 5.0 mmoles of IIIa, 15 mmoles of triethylamine, and 50 ml of anhydrous tetrahydrofuran was treated to the dropwise addition over 15 min of 7.5 mmoles of phosgene in 6 ml of benzene. A solid began to precipitate immediately; after 3 hr of stirring, the crystals were removed by filtration, washed with cold methanol, and recrystallized from dioxane to yield 0.56 g (51%) of white solid, IX, mp 310–312°.

Anal.—Calc. for $C_9H_5N_3O_2S$: C. 49.31; H, 2.30; N, 19.17. Found: C, 49.25; H, 2.42; N, 19.12.

2-Phenyl-3-triazolo[5,1-*b*]benzo[1,3]-thiazin-9-one (X)—The reaction in 65 ml of refluxing dioxane of 5.0 mmoles of IIIa and 10 mmoles of benzoyl chloride after 4 hr of reflux produced an insoluble off-white solid. The reaction mixture was filtered hot to separate the product, X, 0.15 g or 11%, from the unreacted starting materials. Recrystallization from dioxane gave the analytical sample, mp 281–282°.

Anal.—Calc. for $C_{15}H_9N_3OS$: C, 64.50; H, 3.25; N, 15.04. Found: C, 64.26; H, 3.64; N, 14.82.

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[2] Melting points were obtained in capillaries on a Thomas-Hoover apparatus and are uncorrected. IR spectra, in pressed KBr disks, were obtained on a Perkin-Elmer model 257 spectrometer. Combustion analyses were provided by the George I. Robertson Microanalytical Laboratory, Florham Park, N.J.