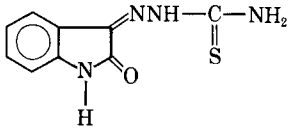
(4) Speiser, P., *A.P.V.-Informationsdienst*, **3**, 53(1964).
(5) Ritschel, W. A., "*Die Tablette*," Aulendorf, 1966, p. 306.
(6) Willstätter, R., *Z. Physiol. Chem.*, **161**, 194, 206 (1926).
(7) Kunitz, M., *J. Gen. Physiol.*, **30**, 291(1947).
(8) Bergmann, M., Fruton, J. S., and Pollok, H., *J. Biol. Chem.*, **127**, 643(1939).
(9) Kwan, K. C., and Milosovich, G., *J. Pharm. Sci.*, **55**, 340(1966).
(10) "Die Substitutionstherapie mit Verdauungsfermenten," *Pharm. Ztg.*, **106**, 594(1961).
(11) Ritschel, W. A., and Orth, H., *Pharm. Ind.*, **28**, 454 (1966).
(12) Anson, M. L., *J. Gen. Physiol.*, **22**, 79(1939).
(13) Lazo-Wasem, E. A., *J. Pharm. Sci.*, **50**, 999(1961).
(14) "Bestimmung der Dextrinierungs-Einheiten," Schweizerische Ferment AG., Basle, Switzerland.

Thiosemicarbazones Derived from Indanedione-1,3

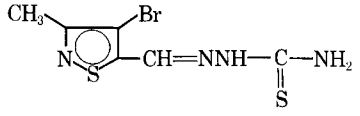
By RAJENDRA S. VARMA and W. LEWIS NOBLES

A series of thiosemicarbazones derived from indanedione-1,3 has been synthesized. Preliminary biological screening data are provided.

ANTIVIRAL (1–7) and tuberculostatic (8–14) activity has been reported for thiosemicarbazones of different carbonyl compounds. Further work on the thiosemicarbazones led to the discovery of the antiviral activity of isatin-3-thiosemicarbazone (I) against the pox group of viruses in human and type 2 polio in ERK cells (15). A number of monocyclic thiosemicarbazones derived from nicotinaldehyde, isonicotinaldehyde, and 2- and 3-thenaldehydes have also exhibited high antiviral activity (5). 4-Bromo-3-methylisothiazole-5-carboxaldehyde thiosemicarbazone (II), when given orally, was found to protect mice infected intracerebrally with neurovaccinia (15). Thiosemicarbazones derived from substituted pyrrolidine-2,3-diones demonstrated protection against experimental influenza infection in mice (15).



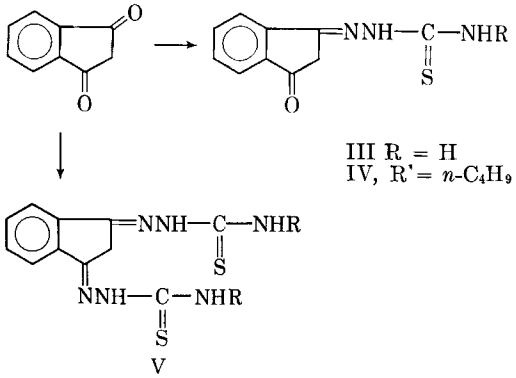
I



II

In view of the antiviral, antituberculous, and antitumor (16, 17) activity demonstrated by certain thiosemicarbazones, the synthesis and biological evaluation of compounds III, IV, and V appeared to be of interest (Scheme I).

Biological Evaluation—Several of the compounds described in this report have been subjected to preliminary antiviral and antibacterial screening procedures. The method with respect to the antiviral screening involved the use of the plastic panel and agar diffusion techniques and, in general, the



III R = H
IV, R' = n-C_4H_9

Scheme I

compounds were screened for possible activity against poliomyelitis type II, *Herpes simplex*, measles, and parainfluenza 3(HA-1) viruses.

In connection with the antibacterial screening, filter paper disks (6.35 mm.) saturated with 2 drops of a suspension of the test compounds (20 mg./ml. in alcohol or water) were tested with various organisms by the use of the agar diffusion method.

Indanedione-1,3-*n*-butylthiosemicarbazone demonstrated activity against the poliomyelitis type II virus as did the indanedione-1,3-methyl dithiosemicarbazone. The other compounds were inactive in this preliminary antiviral screen.

Indanedione-1,3-allyldithiosemicarbazone demonstrated a zone inhibition of 7.5 mm. diameter in the antibacterial screening against *Klebsiella pneumoniae* ATCC 8052; the other compounds reported were inactive against a variety of Gram-positive and Gram-negative organisms utilized in this *in vitro* screening procedure.

EXPERIMENTAL[1]

Indanedione-1,3-thiosemicarbazone (III)—Indanedione-1,3 (0.05 mole) was dissolved in 50 ml. of hot ethanol. To this solution a slurry of thiosemicarbazide (0.05 mole) in 50 ml. of ethanol was added. Instantaneous condensation occurred resulting in the separation of a yellow solid. The reaction mixture was heated for 10 min. and allowed to cool to room temperature. The product was removed by filtration and crystallized from dimethyl-

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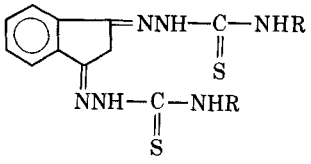
[1] All melting points were taken in capillary tubes on a Thomas-Hoover melting point apparatus and are uncorrected. The infrared spectra were determined using a Perkin-Elmer 137 model spectrophotometer.

TABLE I—INDANEDIONE-1,3-DITHIOSEMICARBAZONES (V)



No.	R	M.p., °C.	Yield, %	Mol Formula	Anal., % Calcd.	Found
1	Methyl	263	78	$C_{13}H_{16}N_6S_2$	C, 48.73	49.15
					H, 5.03	5.17
					S, 20.02	19.79
2	*n*-Butyl	244–245	89	$C_{19}H_{28}N_6S_2$	C, 56.40	56.52
					H, 6.97	6.94
					N, 20.77	19.94
3	Ethyl	245	51	$C_{15}H_{20}N_6S_2$	C, 51.68	52.00
					H, 5.78	5.93
					N, 24.12	23.98
4	Allyl	237	94	$C_{17}H_{20}N_6S_2$	C, 54.80	54.80
					H, 5.41	5.44
					N, 22.56	22.76
					S, 17.22	17.00

formamide–water. Yield 10.5 Gm. (96%), m.p. 230–232° dec. $\nu_{max.}^{KBr}$ 5.85 μ (carbonyl).

Anal.—Calcd. for: $C_{10}H_9N_3OS$: C, 54.79; H, 4.13; N, 19.17. Found: C, 54.82; H, 4.19; N, 19.03.

Indanedione-1,3-n-butylthiosemicarbazone (IV)—To a boiling solution of 4-*n*-butylthiosemicarbazide (0.02 mole) in 25 ml. of ethanol was added indanedione (0.02 mole) dissolved in 25 ml. of ethanol. The resulting reaction mixture was refluxed for 20 min. A solid product separated on cooling; this was suspended in 100 ml. of ethanol–acetone mixture (1:1). The suspension was stirred and filtered. The filtrate, upon refrigeration, yielded a crystalline solid melting in the range of 173–174°. Yield, 5.0 Gm. (91%). $\nu_{max.}^{Nujol}$ 5.88 μ (carbonyl).

Anal.—Calcd. for: $C_{14}H_{17}N_3OS$: C, 61.07; H, 6.22; N, 15.26. Found: C, 61.11; H, 6.14; N, 15.10.

Indanedione-1,3-dithiosemicarbazones—The appropriate thiosemicarbazide (0.15 mole) was dissolved in 50 ml. of ethanol; to this there was added indanedione (0.05 mole). The resulting reaction mixture was refluxed for 4 hr. At the end of this period, the contents were cooled and the product removed by filtration and washed with ethanol. The thiosemicarbazones were recrystallized either from ethyl-acetate or ethanol–dimethylformamide–water. The analyses, melting points, and other pertinent data are recorded in Table I.

REFERENCES

(1) Sadler, P. W., O'Sullivan, D. G., and Bauer, D. J., *Antibiot. Chemotherapy*, **2**, 403(1963).
(2) Bauer, D. J., and Sadler, P. W., *Brit. J. Pharmacol.*, **15**, 101(1960).
(3) Buttimore, D., Jones, D. H., Slack, R., and Wooldridge, K. R. H., *J. Chem. Soc.*, **1963**, 2032.
(4) Slack, R., Wooldridge, K. R. H., McFadzean, J. A., and Squires, S., *Nature*, **204**, 587(1964).
(5) Thompson, R. L., Minton, S. A., Officer, J. E., and Hitchings, G. H., *J. Immunol.*, **70**, 229(1953).
(6) Campaigne, E., Thompson, R. L., and Van Werth, J. E., *J. Med. Pharm. Chem.*, **1**, 577(1959).
(7) Hamre, D., Bernstein, J., and Donovick, R., *Proc. Soc. Expll. Biol. Med.*, **73**, 275(1950).
(8) Domagk, G., *Naturwissenschaften*, **33**, 315(1946).
(9) Behnisch, R., Mietzsch, F., and Schmidt, H., *Angew. Chem.*, **60**, 113(1948).
(10) Buu-Hoi, N. P., Welsch, M., Dechamps, G., Bihan, H. L., Binon, F., and Xuong, N. D., *J. Org. Chem.*, **18**, 121 (1953).
(11) Varma, R. S., *J. Indian Chem. Soc.*, **43**, 558(1966).
(12) Nobles, W. L., *J. Am. Chem. Soc.*, **77**, 6675(1955).
(13) Caldwell, H. C., and Nobles, W. L., *J. Am. Pharm. Assoc., Sci. Ed.*, **45**, 729(1956).
(14) Dodgen, D. F., Jr., and Nobles, W. L., *ibid.*, **46**, 437 (1957).
(15) Whipple, H. E., *Ann. N.Y. Acad. Sci.*, **130**, 71(1965).
(16) Petering, H. G., Buskirk, H. H., and Underwood, G. E., *Cancer Res.*, **24**, 367(1964).
(17) Petering, H. G., VanGiessen, G. J., Crim, J. A., and Buskirk, H. H., *J. Med. Chem.*, **9**, 420(1966).