the stock solutions into 10 ml of sterile nutrient agar to obtain a 10^3 dilution and continuing in the same manner for dilutions up to 10^6. The agar was poured into Petri dishes, allowed to harden, and spot inoculated with 1 drop of a cell suspension of *S. aureus* which was prepared by suspending the growth from a 24-hr nutrient agar slant culture in 10 ml of distilled water. The plates were incubated at 37° for 48 hr and examined for the presence or absence of growth. The results reported in Table I are the minimum concentration of the test compound which will completely inhibit the growth of the bacteria.

Acknowledgment.—We wish to thank Mr. K. L. Godfrey for helpful discussions and advice during the course of the investigation, Mr. P. D. McDonald for the microbiological evaluations, and Messrs. John L. O'Sullivan and Ottmar S. Kring for part of the analytical data.

Derivatives of 5-Carboxymethylthiazolidine-2,4-dione, a New Group of Antiviral Compounds

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Received January 4, 1966

Among the many types of organic compounds which have been tested for their antiviral action, thiosemicarbazones of different carbonyl compounds have been found to be active.[1] Most thiosemicarbazones with high activity contain the group $=NNHCSNH_2$ separated by two carbon atoms from a nitrogen or a sulfur atom. We have recently established that the incorporation of the thiosemicarbazone group in a cyclic system also leads to considerable antiviral activity. Thus 3-(4-bromophenyl)-5-carboxymethylthiazolidine-2,4-dione 2-benzylidenehydrazone was found to prevent at 2×10^{-4} *M* the cytopathogenic changes in cell cultures of human embryonic kidneys infected with herpes simplex virus and poliovirus type 1.[2] Now, some new compounds of this type have been prepared in order to examine some structural effects on their activity. The corresponding thiosemicarbazones of some carbonyl compounds were treated with maleic anhydride in benzene or toluene as described before.[3]

Biological Results.—The inhibitory effect of the compounds under investigation on viral growth as well as the prevention of cytopathogenic changes of the infected cells were tested using human embryonic kidney cell monolayers. The cells were infected with tenfold dilutions of the appropriate virus (from 10^{-3} to 10^{-7}). The test compounds were added usually to the culture medium simultaneously. The concentration of the screened compounds in the culture medium was 5×10^{-5} *M*. At the same time the virus was titrated in the culture medium which contained none of the tested compounds.

Thus 3-(4-bromophenyl)-5-carboxymethylthiazolidine-2,4-dione 2-benzylidenehydrazone delayed the cytopathogenic changes produced by herpes simplex virus strain Z and polio virus no. 1 (Brunhilde) when applied to the infected cells by direct contact or even short time intervals (up to 8 hr) after the infection (therapeutic effect). The virus concentration was lowered on the fifth day for 1.3–2.0 log (virus/ml). Similar results were obtained with polio virus type 2 (Saukett) and vaccinia virus. However, it was inactive with measles virus. All other compounds were tested with herpes virus strain Z and found to reduce the virus titer. The most significant effects were presented by the compounds, collected in Table I.

TABLE I

ANTIVIRAL ACTIVITY

Compd	Reduction of virus concn, log (virus/ml)
1	1.5
3	4.1
5	1.1
8	1.1
9	0.8

Experimental Section

Melting points were determined on a Kofler heating microscope. Ultraviolet spectra were measured with a Beckman Model DU spectrophotometer.

Thiosemicarbazones.—The following compounds were prepared as described in the literature: acetophenone thiosemicarbazone,[4] acetophenone 4-*p*-tolylthiosemicarbazone,[5] propiophenone thiosemicarbazone,[4b,d,6] *p*-dimethylaminobenzaldehyde thiosemicarbazone,[4d,7] furfural thiosemicarbazone,[4b,8] and furfural 4-*p*-tolylthiosemicarbazone.[9] The remainder were obtained by refluxing equimolecular amounts of the corresponding thiosemicarbazide and the carbonyl compound (0.01 mole) in an ethanolic solution for 1 hr and evaporating the solvent *in vacuo* to half of its volume. The crystals thus obtained were crystallized from ethanol. New compounds are as follows.

Acetophenone 4-phenylthiosemicarbazone, mp 193–194°, yield 68%.

Anal. Calcd for $C_{15}H_{15}N_3S$: C, 66.90; H, 5.61; N, 15.61. Found: C, 66.76; H, 5.52; N, 15.78.

Acetophenone 4-*p*-methoxyphenylthiosemicarbazone, mp 183–184°, yield 71%.

Anal. Calcd for $C_{16}H_{17}N_3OS$: C, 64.20; H, 5.72; N, 14.04. Found: C, 64.08; H, 5.65; N, 14.22.

Propiophenone 4-*p*-tolylthiosemicarbazone, mp 92–93°, yield 62%. The product separated as an oil but after 2–4 days crystals were formed.

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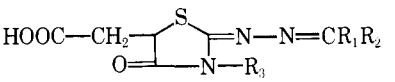
TABLE II
PROPERTIES OF COMPOUNDS PREPARED



Compd	R_1	R_2	R_3	Yield, %	Mp, °C	Formula	Calcd, % C	H	N	S	Found, % C	H	N	S
1[a]	C_6H_5	CH_3	H	80	248–249	$C_{13}H_{13}N_3O_3S$	53.61	4.50	14.43	10.98	53.82	4.76	14.16	11.16
2[b]	C_6H_5	CH_3	C_6H_5	76	188–189	$C_{19}H_{17}N_3O_3S$	62.11	4.66	11.44	8.70	61.85	4.62	11.26	8.62
3[c]	C_6H_5	CH_3	p-$CH_3C_6H_4$	88	219–220	$C_{20}H_{19}N_3O_3S$	62.98	5.02	11.02	8.38	62.96	5.24	10.73	8.56
4	C_6H_5	CH_3	p-$CH_3OC_6H_4$	86	216–218	$C_{20}H_{19}N_3O_4S$	60.45	4.82	10.58	8.05	60.20	4.86	10.68	8.38
5[d]	C_6H_5	C_2H_5	H	81	228–229	$C_{14}H_{15}N_3O_3S$	55.08	4.45	13.77	10.48	55.30	4.77	13.92	10.04
6	C_6H_5	C_2H_5	p-$CH_3C_6H_4$	82	179	$C_{20}H_{21}N_3O_3S$	62.65	5.52	10.96	8.43	62.87	5.49	10.45	8.83
7	p-$(CH_3)_2NC_6H_4$	H	H	78	263–264	$C_{14}H_{16}N_4O_3S$	52.42	5.04	17.49	9.99	52.45	5.14	17.32	10.25
8	2-Furyl	H	H	85	250	$C_{10}H_9N_3O_4S$	44.95	3.40	15.73	11.98	44.76	3.48	15.50	12.31
9	2-Furyl	H	p-$CH_3C_6H_4$	76	230	$C_{17}H_{15}N_3O_4S$	57.14	4.23	11.76	8.96	57.31	4.56	12.17	8.96

[a] λ_{max}^{EtOH} 294 mμ (ε 19,400). [b] λ_{max}^{EtOH} 295 mμ (ε 19,300). [c] λ_{max}^{EtOH} 296 mμ (ε 18,350). [d] λ_{max}^{EtOH} 295 mμ (ε 18,350).

Anal. Calcd for $C_{17}H_{19}N_3S$: C, 68.66; H, 6.44; N, 14.13. Found: C, 68.45; H, 6.38; N, 14.20.

Derivatives of 5-Carboxymethylthiazolidine-2,4-dione. General Procedure.—Equimolecular amounts of the corresponding thiosemicarbazones and maleic anhydride (usually 0.01 mole) were suspended in 40 ml of benzene (or toluene), and the mixture was refluxed for 2 hr. Upon cooling to room temperature, the product was filtered off, washed with the solvent employed in the reaction, dried, and crystallized from ethanol. The compounds with the corresponding analytical data are presented in Table II.

3-Aryl-5-halomethylisoxazoles. A New Class of Anthelmintics

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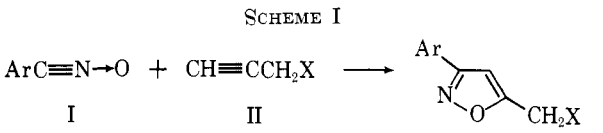
Isoxazoles have been reported to possess diverse biological activities[1] but to date there is no reference, in the literature, to any isoxazole having anthelmintic activity. In our continuing efforts to find suitable drugs for tapeworm, pinworm, and hookworm infections, we had occasion to synthesize 4-bromomethyl-3-(4-chlorophenyl)isoxazole (**1**, Table I) and subject it to anthelmintic screening. While it was devoid of any activity against oxyurids and *Nematospiroides* infections in mice, it exhibited pronounced activity against *Hymenolepis nana* infection when administered orally. Accordingly, 14 analogs of this compound (see Table I) were prepared and examined for anthelmintic activity.

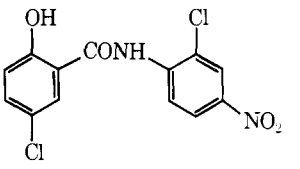
Chemistry.—The 3-aryl-5-bromomethylisoxazoles (IIIc) were prepared by the dipolar cycloaddition of various benzonitrile oxides (I) to propargyl bromide (II, X = Br) essentially according to the procedure of D'Alcontres and Lo Vecchio.[2] These could be converted smoothly into the corresponding 5-iodomethyl derivatives (IIId) by treatment with potassium iodide in anhydrous dimethylformamide. The 5-chloromethyl analogs (IIIb) were obtained by the action of thionyl chloride on the respective 5-hydroxymethyl derivatives (IIIa) which were in turn prepared by treating benzonitrile oxides (I) with propargyl alcohol (II, X = OH).[3] Compounds of the type IIIb can also be prepared directly by the cycloaddition of benzonitrile oxides (I) to propargyl chloride (II, X = Cl)[4] (see Scheme I).

SCHEME I



I II

IIIa, X = OH
b, X = Cl
c, X = Br
d, X = I



IV

Biology.—Swiss albino mice of either sex weighing approximately 20 g each were used in all the experiments. Each animal was experimentally induced to *H. nana* and *N. dubius* infections. Oxyurid infections due to *Syphacia obvelata* and *Aspicularis tetraptera* occurred naturally.

Viable *H. nana* eggs were obtained from the gravid segments of worms harbored by the untreated mice and counted in three 0.1-ml samples following the technique of Standen.[5] Infective *N. dubius* larvae were obtained according to the method of Sheffield, *et al.*[6] A suspension of these larvae was mixed with one of *H. nana* eggs in such proportions as to furnish a final mixture with concentrations of 250–300 larvae/ml and 5000–6000 eggs/ml. Each mouse was administered 0.2 ml of this suspension by gavage.

The infected mice were randomly divided into treated and control groups. On the 19th or 20th day postinfection, the mice in the test groups were treated

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