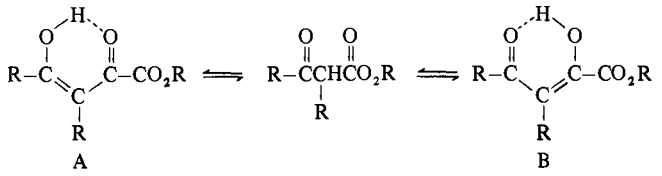

A B

growth of both organisms. A series of compounds was then prepared by known methods in order to investigate possible structure-activity relationships. Table I summarizes the physical and biologic properties of these compounds.

All of the chlorinated analogs (**14-20, 22**), prepared by SO_2Cl_2 chlorination of the respective acylpyruvates, were strong vesicants. Except for **22**, these compounds were the most active antifungal compounds. Lengthening of the chain from 5 to 9 C atoms (**1, 5-8**) resulted in diminished activity. However, increasing the C chain length of the ester group (**1-4**) had no significant effect on activity. Hydrolysis of ester **1** to acetopyruvic acid (**21**) resulted in the loss of antifungal activity at the drug level tested. The fact that the 3,3-dichloro ester **22** was inactive at such levels, whereas the 3-chloro ester **15** and the unchlorinated ester **1** were active, suggests that the formation of the enolic structure A or B may be a prerequisite for antifungal activity.

Ester **20** at 10 μg/ml or less inhibited growth in a minimal inhibitory concn test against the following additional species of yeasts: *Candida tropicalis, C. krusei, C. guilliermondi,* and *Torulopsis glabrata.* Ester **20**, in a toxic agar test, showed complete inhibition of *Aspergillus niger* at 10 μg/ml of medium.

Experimental Section

Melting points were determined in a Mel-Temp capillary melting point apparatus and are corrected. Boiling points are uncorrected.

Acylpyruvates. The general procedure of Royals[3] was used to prepare the esters starting with the appropriate Me ketone and dialkyl oxalate.

Ethyl 3,3-Dichloro-2,4-dioxovalerate (22). To 158 g (1 mole) of 1 cooled to 0° was added dropwise below 10° with stirring 270 g (2 moles) of freshly distd SO_2Cl_2. The addition required 1 hr. Stirring was then continued at 25° for 4 hr. Gases (SO_2 and HCl) were removed *in vacuo* for 0.5 hr. To the residue was added 200 ml of H_2O and 100 ml of $CHCl_3$. The mixture was shaken thoroughly and the layers were separated. The $CHCl_3$ layer was washed with 200 ml of H_2O in 2 portions. The combined aqueous layer and washings were extracted once with 50 ml of $CHCl_3$. The combined exts were dried ($MgSO_4$) and the solvents were distd at atm pressure through a Claisen head to give 100 g of **22**; bp 128-130°. Redistillation through a 457 cm Vigreux column gave a 76% recovery of **22** as a colorless, pungent oil.

The yellow monochloro compds, **14-20**, were prepd by a similar procedure using only 1 equiv of SO_2Cl_2. $CHCl_3$ could be used as a solvent without lowering the yields.

Acknowledgments. The authors are indebted to Grant Gustin and Marvin Tefft for the elemental analyses, and to Warren Smith, Ralph Bush, and June Horton for technical assistance during the preparation and testing of the compounds.

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Biologically Oriented Organic Sulfur Chemistry. 10. Inhibitory Effects of Certain Organic Sulfur Compounds on *Histoplasma capsulatum*[1,†]

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Preliminary study of several classes of organic S compds gave promising leads to possible chemotherapeutic agents for the inhibition of *Histoplasma capsulatum.*[2,‡] This suggested, as a guide to more intensive study of the most attractive classes, that it would be wise to examine as many different classes of organic S compds as were available to us. Previous results have been reported.[1,4] This paper continues the study with some 13 different classes of organic S compds comprising 61 representatives. Table I shows the results of *in vitro* tests, carried out as described previously,[2,4a,4c] for inhibition of growth of the yeast phase of *H. capsulatum.*

Certain thiols have shown promise,[4a] but of the group **1-16** only **1** and **2** were significantly active, in common with their resemblance to *p*-chlorobenzenethiol which was active at 2.5-7.5 μg/ml.[4a] The low activities of **3-5** are consistent with an earlier conclusion that, although electron-withdrawing substituents seem helpful (as with **1**), too effective a withdrawal seems deleterious.[4a] The inactivity of **6** is surprising, in view of the similarity to *p*-chlorobenzenethiol. Since 2-naphthalenethiol was promising (~5 μg/ml),[4a] the reduced activity of the 1 analog (**7**) is interesting. Also interesting is the inactivity of **8**, since acetyl allyl disulfide was active (5-10 μ/ml);[4d] thioacetic acid is not very active (13-15 μg/ml),[4a] so perhaps a hydrodisulfide moiety (RSSH) actually is the effective agent with the disulfide. The low activity of **9** is not surprising, since electron donation seems to decrease activity.[4a] Inactivity of **10** parallels that of *o*-mercaptobenzoic acid.[4a]

The apparent difference in activity of diacetyl sulfide **17** and diacetyl disulfide **18** is intriguing, since it seems likely that **17** and **18** would have similar pharmacological properties of transport and the like. The fact that **18**, unlike **17**, can produce a hydrodisulfide supports the suggestion already made of the possible functioning of a hydrodisulfide

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‡Also, fungistatic activity of certain organic sulfur compds was reported by Buckman, *et al.*[3]

Table I. Inhibitory Effects of Organic Sulfur Compounds on the Growth of *Histoplasma capsulatum*

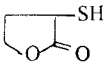
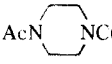
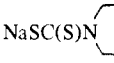
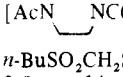
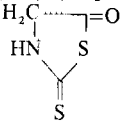
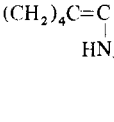
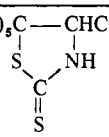
Class	Compd[a]	Structure[b]	MIC, μg/ml[c,d] H-7	MIC, μg/ml[c,d] H-25	Solvent[e]
Thiols	1[f]	4-BrPhSH	10	5	MT
	2[f]	3,4-Cl_2PhSH	15 (p10)	5	MT
	3[f]	Cl_3CSH	...	...	DT
	4[f]	2-C_5H_4N-SH	...	p20	MW
	5[f]	4-C_5H_4N-SH	15	15	MT
	6[f]	ClPh-2,4-$(SH)_2$	...	...	MT
	7[f]	1-$C_{10}H_7SH$	20 (p15)	10	MT
	8[f]	H_2C=$CHCH_2SH$	...	...	MT
	9[f]	4-$HSPhNH_2$	p20	20	MT
	10	2-HSPh-1,4-$(CO_2H)_2$	...	...	W[g]
	11[f]		...	...	MT
	12	$HS(CH_2)_3CO_2H$	...	...	MW
	13	$HS(CH_2)_4CO_2H$	...	...	MW
	14[f]	2-$C_{10}H_7NHC(O)CH_2SH$	...	10	MT
	15[f]	2-SH-benzimidazole	...	...	MT*
	16[f]	6-SH-purine	...	...	MT*
Sulfides	17	$(Ac)_2S$	10	15	MT
Disulfides	18	$(AcS)_2$	10 (p5)	5	MT
	19	$AcSSCH_2Ac$[h]	10	10	MT
	20	$AcSSPhCl_5$	20	20	MT[i*]
	21[f]	(2-$C_5H_4NS)_2 \cdot H_2O$	10	10	MT
	22[f]	$[SC(CH_3)_2CH(NH_2)CO_2H]_2$	...	...	MT
	23	2-HO_2CPhSSEt	...	...	MT
	24	2-HO_2CPhSS-*tert*-Bu	...	...	MT*
	25	2-HO_2CPhSS-*trans*-2-Cl-cyclohexyl	...	...	MT
	26	2-$HO_2CPhSSCH_2CH(NH_2)CO_2H$	...	...	MT*
	27	2-HO_2CPhSSPh	...	...	DW*
	28	2-HO_2CPhSSPh-4-Cl	...	...	MT
	29	2,5-$(H_2NCH_2CH_2SS)_2$Ph-1,4-$(CO_2H)_2$	...	...	W[g]
	30	2-$NH_2(CH_2)_2SSPh$-1,4-$(CO_2H)_2$	...	...	W[g]
	31	2,5-(*n*-$C_{10}H_{21}NHCH_2CH_2SS)_2$Ph-1,4-$(CO_2H)_2$	p20	10	W[g,i*]
Dithiocarbamates	32		...	...	W
	33		...	...	W
	34		...	...	W
	35	2,5-$[Me_2NC(S)S]_2$Ph-1,4-$(CO_2Et)_2$	...	...	W[i]
	36	$(CH_2)_5NC(S)S(CH_2)_2SS(CH_2)_2NHAc$	...	p20	MT
	37	$[(CH_2)_5NC(S)S(CH_2)_2S]_2$	...	...	MT[i*]
Trithiopercarbamates and thiuram sulfides	38	$(CH_2)_5NC(S)SS$-*tert*-Bu	p15	20	MT
	39[f]	$[(n\text{-}Bu)_2NC(S)]_2S$	...	...	MT*
	40		...	...	MT*
Thiocyanates	41[j]	*n*-$BuSO_2CH_2SCN$	15	15	DT
	42[j]	2-Benzothiazolyl-SCH_2SCN	10 (p5)	5 (p2.5)	DT
	43[j]	$CH_2(SCN)_2$	2.5	2.5	DT*
Thiolsulfonates	44[j]	*n*-$BuSO_2SCH_2Cl$	15	10	MT
	45[j]	$MeSO_2SCH_2CH(OH)CH_3$	...	...	W
	46[j]	$CH_2(SSO_2$-*n*-$Bu)_2$	...	...	DT
Sulfones	47	$PhSO_2CH$=$CHPh$	...	...	DT
	48	$PhSO_2CH_2CH(OH)Ph$	...	...	W
	49	$PhSO_2CH_2CHClPh$	...	p20	MT
	50	*p*-$C_7H_7SO_2CH_2COPh$	...	...	MT*
	51	*p*-$C_7H_7SO_2CH_2C$(=NH)Ph	...	p20	MT
	52	$PhSO_2CH_2C$(=NNHPh)Ph	...	...	MT*
	53	*p*-$C_7H_7SO_2C(CH_3)_2C(CH_3)_2OH$	...	...	MT
	54	*p*-$C_7H_7SO_2CH(CONHPh)_2$	...	...	*k*
	55	*p*-$C_7H_7SO_2(CH_2)_3OC(O)$-3,5-$(NO_2)_2$Ph	...	...	DT[i*]
2-Thiazolidinethiones	56		10	10	MT
	57		...	...	DT*

Table I *(Continued)*

Class	Compd[a]	Structure[b]	MIC, μg/ml[c,d] H-7	MIC, μg/ml[c,d] H-25	Solvent[e]
	58		–	–	MT*
Miscellaneous	59[f]	$CH_3C(O)SMe$	–	–	MT
	60	$CH_3C(S)OEt$	–	–	MT
	61	p-$C_7H_7SO_2S^-K^+$	–	–	W

[a]Compds were prepared and suitably characterized (except for 19) in our laboratories by methods published elsewhere, except where other sources are indicated. [b]C_5H_4N = pyridyl; $C_{10}H_7$ = naphthyl; p-C_7H_7 = p-tolyl. [c]MIC = Minimum inhibitory concn. Numerals indicate the lowest concn in μg/ml that resulted in complete inhibition of growth during a 7-day incubation period. A numeral preceded by the letter "p" indicates that inhibition was partial at the concn indicated;— means no inhibition was evident after 7 days of incubation. General methods and strains H-7 and H-25 were as described in ref 2, 4a, and 4c. [d]The highest concn tested was 20 μg/ml except for **28** and **57** where it was 15 and 7.5 μg/ml, respectively. [e]Compds dissolved in dioxane (D) or MeOH (M) were dild with a 0.04% aqueous soln of Tween 80 (T) or water (W) so that the following final concns in the medium were not exceeded: M, 0.25%; T, 0.008%. W = soln in H_2O. An asterisk indicates that the compd did not dissolve completely. Hot solvent was used to dissolve **1**, **10**, **14**, **30**, **40**, **43**, **45–48**, **52**, and **57**. [f]Commercial sample. [g]NaOH added to effect soln. [h]Efforts to purify **19**, described in ref 4d, were unsuccessful, and the assay had to be done with crude **19**. [i]The samples **20** (in MT), **31** (in 0.01 *N* NaOH), **35** (in W), **55** (in DT), and **37** (in the medium) were sterilized in an autoclave; some destruction of the compd may have resulted. [j]Compd kindly provided by Dr. J. D. Buckman, Buckman Laboratories, Inc., Memphis, Tenn. [k]Compd **54** was dissolved in alcoholic NaOH, which then was brought as near neutrality as possible (using AcOH) without causing pptn.

moiety. Among the disulfides **18–31**, only **18–21** and **31** showed much activity. Compds **19** and **20** might be expected to produce hydrodisulfides. Acyl disulfides with structures resembling **18–20** previously were reported to be active.[4c] The activity of **21** is of interest since the corresponding thiol **4** was virtually inactive. The activity of **31** may result from decomposition to 2-(*n*-decylamino)ethanethiol (MIC 7.5–10 μg/ml).[4a]

Certain dithiocarbamates, trithiopercarbamates, and thiuram disulfides have proved promising as inhibitors of *H. capsulatum*.§ However, the dithiocarbamates **32–37** were inactive or nearly so, as were the trithiopercarbamate **38** and the thiuram-type structures **39** and **40**. Perhaps the significance is that relative simplicity is the keynote, so that bulky or polar types of molecules exemplified by **32–40** are unsatisfactory because of poor transport through membranes or because of adsorption at improper sites.

The thiocyanates **41–43**, the first we have examined, show quite promising activity and appear to provide a lead that should be pursued.

The thiolsulfonates **44–46** and the sulfones **47–55** (a major class we have not tested heretofore) showed little (**44**, **49**, and **51**) or no activity. The thiazolidine **56** showed reasonable promise, but inactivity of the variations **57** and **58** make further work in this area unattractive; moreover, even **56** was unpromising *in vivo*.#

S compds with low molecular weight usually have given the best results.[4a,c] The simple thiol and thion esters **59** and **60** therefore were logical for trial; neither was active. The thiosulfonate salt **61** was of interest because of the activity of methyl methanethiolsulfonate,[4a] but in common with the thiolsulfonates **44–46** no useful lead developed.

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§For examples and leading references, see ref 2.

#There was no prolongation of the life of mice exposed to X-rays and then infected with *H. capsulatum* before treatment with **56**. We are indebted for this test to Drs. R. S. Gordee and W. B. Lacefield of Eli Lilly and Co.; it was performed essentially by methods described in ref 1 and 4c.

Quinoxaline Studies. 20.[1†] Potential Antimalarials. Synthesis of *anti*- and *syn*-*N,N*-Dialkylaminomethyl 2-Quinoxalinyl Ketoximes

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Unlike many quinolinemethanols,[2] quinoxalinemethanols were demonstrated[3,4] to possess no useful antimalarial activity. It was hoped, however, that aminomethyl 2-quinoxalinyl ketoximes, possessing as they do the seemingly requisite OH and amine functions, would display antimalarial activity. The purpose of this paper is to report the synthesis of a series of *N,N*-dialkylaminomethyl 2-quinoxalinyl ketoximes for testing as antimalarials; neither oximes nor intermediates, unfortunately, had significant antimalarial activity (Table I).[5,‡]

Chloromethyl 2-quinoxalinyl ketone (**1**)[3] was chosen as the starting material for this investigation. Because of previous demonstration[3] of the instability of the corresponding aminomethyl 2-quinoxalinyl ketones, the sequence **1**, chloromethyl 2-quinoxalinyl ketoxime (**2**), and aminomethyl 2-quinoxalinyl ketoxime (**3**) was dictated as the only reasonable course to the objective described in this paper.

2-Chloroacetylquinoxaline formed **2**, a single oxime (nmr), which decomposed when subjected to a variety of Beckmann rearrangement reagents. Examination of Dreiding

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