

Table I.

<div><div><div><div><div><div></div><div>O</div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div><div><div><div><div><div></div><div></div><div></div></div></div></div></div><div><div><div><</div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div></div>							
--	--	--	--	--	--	--	--

^aAll new compds were analyzed for C, H, and N and results are within ±0.4%. ^bLit. mp 58–62. ^cMp varies with rate of heating. ^dLit mp 159–160 dec. ^eDouble mp. ^fRun in CDCl₃. ^gRun in DMSO-*d*₆. ^hRun in CF₃CO₂H-CDCl₃. ⁱThe sample seems to decompose in DMSO-*d*₆ as evidenced by a change in spectrum; e.g., immediate run 7.3–8.2 (m), 11.13 (s), 13.57 (broad m); on standing overnight 7.3–8.3 (nature of m has changed), 13.57 (broad m). ^jRun in D₂O. ^kCalcd C, 80.71; found C, 81.27. ^lCalcd C, 72.62; found C, 71.28.

in H₂O and making basic (to about pH 8) with solid NaHCO₃, followed by standard work-up. See Table I for the results.

References

- (1) P. M. Weintraub, *Chem. Ind. (London)*, 1296 (1970) (part 4).
- (2) E. Ochiai, "Aromatic Amine Oxides," Elsevier Publishing Company, New York, N.Y., 1967, pp 424–442; M. H. Bickel, *Pharm. Rev.*, **21**, 325 (1969).
- (3) (a) D. Diels and D. Riley, *Ber.*, **48**, 897 (1915); (b) W. Dilthey and J. Friedrichsen, *J. Prakt. Chem.*, **127**, 292 (1930); (c) J. W. Cornforth and R. H. Cornforth, *J. Chem. Soc.*, 96 (1947); (d) C. M. Selwitz and A. I. Kosak, *J. Amer. Chem. Soc.*, **77**, 5370 (1955); (e) K. Bodendorf and H. Towliati, *Arch. Pharm. (Weinheim)*, **298**, 293 (1965).

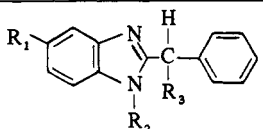
Antiviral Agents. 2. Analogs of 2-(α-Hydroxybenzyl)benzimidazole

F. Gualtieri,† G. Brody, A. H. Fieldsteel, and W. A. Skinner*
Life Sciences Division, Stanford Research Institute, Menlo Park, California. Received August 19, 1971

A large number of analogs of the antiviral agent, 2-(α-hydroxybenzyl)benzimidazole (HBB), have been synthesized and evaluated for their effects on various viruses. One of the more recent publications¹ reported on the antiviral

†Postdoctoral, NATO Fellow, SRI.

Table I.^a Analogs of HBB

Compound	R ₁	R ₂	R ₃	Mp, °C	Recryst solvent ^b	Yield, %	Formula ^c
							
1	H	H	Cl	158-160	A	75	^d
2	H	H	SH	233-234	B	49	C ₁₄ H ₁₂ N ₂ S
3	H	H	NH ₂	201-202	C	49	^e
4	H	H	OCH ₃	159-161	D	83	^f
5	H	H	OC ₂ H ₅	209-210	E	73	C ₁₆ H ₁₆ N ₂ O
6	H	H	OC ₃ H ₇	216-217	E	73	C ₁₇ H ₁₈ N ₂ O
7	H	H	NHCH ₃	158-160	C	72	C ₁₅ H ₁₅ N ₃
8	H	H	N(CH ₃) ₂	202-204	E	90	C ₁₆ H ₁₇ N ₃
9	H	H	NHCOCH ₃	251-252	C	24	C ₁₆ H ₁₅ N ₃ O
10	H	COCH ₃	NHCOCH ₃	207-209	F	43	^g
11	Cl ^h	H	SH	211-213	E	15	C ₁₄ H ₁₁ ClN ₂ S
12	Cl ^h	H	OCH ₃	184-185	G	57	C ₁₅ H ₁₃ ClN ₂ O
13	Cl ^h	H	NHCH ₃	178-179	C	62	C ₁₅ H ₁₄ ClN ₃
14	CH ₃ ^h	H	SH	205-207	E	43	C ₁₅ H ₁₄ N ₂ S
15	CH ₃ ^h	H	OCH ₃	166-168	C	70	C ₁₆ H ₁₆ N ₂ O
16	CH ₃ ^h	H	NHCH ₃	152-154	C	74	C ₁₆ H ₁₇ N ₃
17	CH ₃ ^h	H	N(CH ₃) ₂	164-166	E	80	C ₁₇ H ₁₉ N ₃
18	CH ₃ ^h	H	SCH ₃	184-186	E	58	C ₁₆ H ₁₆ N ₂ S
19	H	CH ₃	SH	184-186	C	34	ⁱ
20	H	CH ₃	OCH ₃	72-73	^j	70	C ₁₆ H ₁₆ N ₂ O
21 ^k	H	CH ₃	NHCH ₃	218-220	E	39	C ₁₆ H ₁₇ N ₃ ·Pic
22 ^k	H	CH ₃	N(CH ₃) ₂	170-172	E	77	C ₁₇ H ₁₉ N ₃ ·2Pic
23	H	CH ₃	SCH ₃	78-79	H	51	C ₁₆ H ₁₆ N ₂ S
24	H	C ₆ H ₅	OCH ₃	93-94	H	87	C ₂₁ H ₁₈ N ₂ O
25	H	C ₆ H ₅	NH ₂	88-89	I	25	C ₂₀ H ₁₇ N ₃
26	H	C ₆ H ₅	NHCOCH ₃	216-218	E	79	C ₂₂ H ₁₉ N ₃ O
27	H	C ₆ H ₅	NHCH ₃	143-145	C	54	C ₂₁ H ₁₉ N ₃

^aIr and nmr spectra of the compounds of this table are consistent with the proposed structure. ^bA = acetone; B = AcOH; C = MeNO₂; D = EtOAc; E = EtOH; F = Ac₂O; G = Me₂CO-H₂O; H = hexane; I = PhH-petr ether. ^cAnal. C, H, N (2, 14, 18, 19, and 23 were also analyzed for S). ^dW. R. Siegart and A. R. Day, *J. Amer. Chem. Soc.*, **79**, 4391 (1967). ^eM. Mengelberg, *Chem. Ber.*, **92**, 977 (1959). ^fD. G. O'Sullivan, D. Pantic, and A. K. Wallis, *Nature (London)*, **205**, 262 (1962). ^gAnal. Calcd: C, 70.34; H, 5.58; N, 13.67. Found: C, 69.44; H, 5.64; N, 13.97. ^hTautomeric forms exist, 5 or 6 position. ⁱAnal. Calcd: C, 70.85; H, 5.55; N, 11.01; S, 12.58. Found: C, 70.69; H, 5.05; N, 11.17; S, 12.65. ^jPurified by column chromatog (SiO₂; EtOAc-cyclohexane 20:30 as eluant). ^kOily; identified as picrates.

activity of analogs of HBB against ECHO 6 virus in monkey kidney cells. Of the 42 compounds evaluated, only 2-(α -methyl- α -hydroxybenzyl)benzimidazole was more active and more selective than HBB. The α -ethyl derivative was more active but less selective than HBB.

An earlier report² indicated that with polio viruses, the α -methyl and related derivatives gave less protection of infected cells than did HBB. This is in agreement with the known differences between polio and other enteroviruses in sensitivity to HBB.³ Gwaltney⁴ investigated the activity of HBB against rhinovirus strains representative of the 55 numbered serotypes and one subtype. Half the strains showed inhibition at a 447 μ M concentration of HBB and strains of all types except 1A and subtype 1B showed some degree of inhibition at a concentration of 574 μ M or less.

As a continuation of our studies on structure-activity relations in antiviral agents of the HBB type⁵ we synthesized a group of benzimidazoles as shown in Table I. These compounds were evaluated in tissue culture for their antiviral activity (Table II). Standard reference compounds were previously reported⁵ using this assay method.

The synthesis of these HBB analogs was *via* the 2-(α -chlorobenzyl)benzimidazole which was synthesized from the 2-(α -hydroxybenzyl)benzimidazole with SOCl₂. The active Cl was then replaced with the appropriate grouping to yield the desired 2-(α -substituted benzyl)benzimidazole.

Due to the extreme reactivity of the α -chlorobenzyl compound, it was not purified in most cases prior to the final reaction. The mercapto derivatives were particularly sus-

ceptible to oxidation and in the case of 2 the decomposition product of heating was isolated and identified as 2-benzoylbenzimidazole.

Antiviral Activity. Initially, HBB was evaluated by the method previously reported,⁵ against the following viruses in tissue culture using primary culture of rhesus monkey kidney: ECHO-12, polio 1, vaccinia, influenza A2, vesicular stomatitis virus (VSV), and adenovirus-7. Activity was found only against ECHO-12 and polio 1. Activity was highest against ECHO-12. None of the analogs synthesized were active against polio 1 but some were quite active against ECHO-12.

The most interesting findings in this series of HBB analogs were the activities of the amino analogs of HBB, 2-(α -amino-benzyl)benzimidazole (3) and 1-phenyl-2-(α -aminobenzyl)-benzimidazole (25). Against ECHO-12, 3 gave 100% reduction of the cytopathic effect at 125 μ g/ml test concn with the cytotoxic level at 1000 μ g/ml.

To determine cytotoxicity, each compd is tested to determine the highest concn that will be tolerated by the cell culture without inducing obvious cellular damage. To make this determination, approximately 20 mg of compd is dissolved in an amount of tissue culture medium to give a concn of 500 μ g/ml. If the compd does not go into solution, it is subjected to sonic vibration to produce a homogeneous suspension. Serial twofold dilutions down to 1 μ g/ml are then made, using tissue culture medium as diluent; 2 ml of each dilution serving as sole source of medium is then added to four monkey kidney tubes. The tubes are then placed in a

Table II. Antiviral Activity against ECHO-12 Virus

Compound	Concentration, $\mu\text{g/ml}$			Therapeutic index ^b
	Cytotoxic	Lowest level causing 100% reduction in cytopathic effect of the virus	EC_{50} ^a	
HBB-dl·Base	250	62	28	8.9
HBB-dl·HCl	125	62	27	4.6
HBB-d(-)·HCl	250	31	20	12.5
2	16	No activity		
3	1000	125	72	13.9
4	125	No activity		
5	31	No activity		
6	31	No activity		
7	62	No activity		
8	62	No activity		
9	125	No activity		
10	125	No activity		
11	31	No activity		
12	16	No activity		
13	16	No activity		
14	250	No activity		
15	62	No activity		
16	62	31 ^c		
17	250	No activity		
18	125	No activity		
19	31	No activity		
20	62	No activity		
21	62	31	14	4.4
22	16	No activity		
23	16	No activity		
24	31	8	4	7.7
25	62	4	3	20.7
26	16	No activity		

^aConcn in $\mu\text{g/ml}$ resulting in 50% reduction in cytopathic effect of the virus; estimated using the log probit method.⁸ ^bTherapeutic index is the ratio of the cytotoxic concn to the EC_{50} . ^c100% active at 31 $\mu\text{g/ml}$ when the dose of virus was 20 ID_{50} ; at a virus dose of 200 ID_{50} , only 27% activity was noted.

roller drum at 35° and incubated for 7 days. The cultures are examined microscopically at intervals to determine the highest concn of the compd that is nontoxic.

Compd 25 gave 100% reduction of the cytopathic effect at 4 $\mu\text{g/ml}$ with the cytotoxic level at 62 $\mu\text{g/ml}$. Alkylation or acetylation of the amino function resulted in loss of antiviral activity with the aromatically unsubstituted benzimidazole moiety. The presence of a 5- or 1-Me group in the *N*-methylamino series increased activity against ECHO-12. Substitution in the 1 position by Ph increased the activity against ECHO-12 of the 2-(α -methoxybenzyl)benzimidazole (4) as well as the α -amino derivative (3). Although the concn of the 1-Ph derivative of 4 (24) causing 100% reduction of the cytopathic effect was much lower than HBB, the increased cytotoxicity of the compound resulted in a therapeutic index of 7.7 as compared to 8.9 for HBB. The therapeutic index of 25 was, however, 20.7; considerably better than that of HBB. This compd, in contrast to HBB, was found to be inactive against polio 1.

During the preparation of this manuscript a paper appeared in which the 2-(α -aminobenzyl)benzimidazole was reported⁶ to be active against poliovirus types 1, 2, and 3. However, it was not found to be as effective as HBB in protecting ERK cells against the cytopathic effects of types 1, 2, and 3 polioviruses.

Experimental Section

2-(α -Chlorobenzyl)benzimidazoles. A soln of SOCl_2 (0.06 mole) in 15 ml of CHCl_3 was added with stirring and cooling to the

suitable 2-(α -hydroxybenzyl)benzimidazole (0.02 mole) and the mixt heated to reflux for 4 hr. The soln was evapd to dryness *in vacuo* and the solid washed with anhyd Et_2O . The solid crude product was used for further reactions except in the case of 1-phenyl-(α -chlorobenzyl)benzimidazole hydrochloride, recrystd as the hydrochloride, and 2-(α -chlorobenzyl)benzimidazole which was isolated as the free base.

2-(α -Methoxybenzyl)benzimidazole (4). 2-(α -Chlorobenzyl)benzimidazole (0.01 mole) and NaOMe (0.01 mole) were dissolved in 50 ml of MeOH and left at room temp overnight. The soln was evapd *in vacuo* and the resultant solid was washed with H_2O , dried, and recrystd from EtOAc, mp 159–161°. All other alkoxy deriv were prepd in the same way.

2-(α -Methylaminobenzyl)benzimidazole (7). 2-(α -Chlorobenzyl)benzimidazole (0.01 mole) was dissolved in anhyd dioxane (50 ml) and an excess of MeNH_2 (40% H_2O sol; 10 ml) was added. After standing overnight at room temp, the soln was evapd to dryness and the gummy solid was washed with H_2O , dried, and crystd from MeNO_2 , mp 158–160°. All other alkylamino derivs were obtained in the same manner. Oily derivs were purified by alumina chromatog using EtOAc-cyclohexane (80:20) as elutant. They were characterized as picrates.

2-(α -Methylthiobenzyl)-5(6)-methylbenzimidazole (18). 2-(α -Chlorobenzyl)-5(6)-methylbenzimidazole hydrochloride (0.01 mole) was dissolved in 50 ml of anhyd dioxane and the soln added to 10 ml of a 2 *M* soln of NaSCH_3 . After standing overnight at room temp, the dioxane was removed *in vacuo* and the residue extd with CHCl_3 and dried (Na_2SO_4). The solid obtained upon evaporation was crystd from EtOH- H_2O , mp 184–186°.

2-(α -Mercaptobenzyl)benzimidazole (2). 2-(α -Chlorobenzyl)benzimidazole (0.01 mole) was dissolved in dioxane (20 ml) and the soln added to a soln of 0.04 *M* KOH in 50 ml of H_2O previously satd at 0° with H_2S . After standing overnight at room temp, a solid was pptd, filtered, and washed with H_2O , EtOH, and Et_2O . The solid was crystd from AcOH, mp 233–234°. From the mother liquor a cryst solid was obtained that was identified as 2-benzoylbenzimidazole. Other α -mercaptobenzyl derivatives were obtained in the same way except that occasionally acidification after the reaction was necessary to ppt the product.

2-(α -Aminobenzyl)benzimidazole (3). 2-(α -Chlorobenzyl)benzimidazole (0.01 mole) was dissolved in 50 ml of dioxane and added to 10 ml of concd NH_4OH . After 12 hr at room temp the soln was evapd to dryness and the solid extd with 2 *M* HCl. The acidic ext was made alk with 2 *N* NH_4OH and a solid was obtained which was washed with H_2O and dried *in vacuo*. Purification was by column chromatog (Al_2O_3) using EtOAc-cyclohexane (1:1) as eluant. Crystn was from MeNO_2 , mp 201–203° (lit.⁷ mp 202–203°).

2-(α -Acetamidobenzyl)benzimidazole (9). 2-(α -Aminobenzyl)benzimidazole (3) (0.5 g) was dissolved in Ac_2O and left at room temp for 48 hr. A white solid was obtained that could be crystd from Ac_2O . This was found to be the extremely unstable diacetyl deriv, hydrolyzing to the mono-Ac deriv readily. This compd can be crystd from MeNO_2 , mp 251–252°.

Acknowledgments. The authors wish to thank J. Sorge, K. Young, and C. Kurahara for technical assistance in carrying out the antiviral experiments and M. Leaffer for assistance in the synthetic portion. The financial assistance of the SRI-Institute Research and Development Program is acknowledged for the antiviral studies.

References

- (1) I. Tamm, H. J. Eggers, R. Bablanian, A. F. Wagner, and K. Folkers, *Nature (London)*, **223**, 785 (1969).
- (2) D. G. O'Sullivan and A. K. Wallis, *ibid.*, **198**, 1270 (1963).
- (3) I. Tamm and H. J. Eggers, *Virology*, **18**, 439 (1962).
- (4) J. M. Gwaltney, Jr., *Proc. Soc. Exp. Biol. Med.*, **129**, 665 (1968).
- (5) F. Gualtieri, G. Brody, A. H. Fieldsteel, and W. A. Skinner, *J. Med. Chem.*, **14**, 546 (1971).
- (6) D. G. O'Sullivan, C. Ludlow, D. Pontic, and A. K. Wallis, *Antimicrob. Ag. Chemoth.*, **153** (1969).
- (7) M. Mengelberg, *Chem. Ber.*, **92**, 977 (1959).
- (8) L. C. Miller and M. L. Tainter, *Proc. Soc. Exp. Biol. Med.*, **57**, 261 (1944).