A New Antifungal and Antibacterial Agent, Methyl 5(*or* 4)-(3,3-dimethyl-1-triazeno)imidazole-4(*or* 5)-carboxylate

Sir:

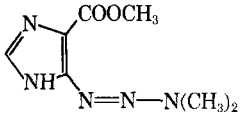
The relative scarcity (1–3) of useful antifungal drugs in comparison with the many antibacterial agents of microbial and synthetic origins emphasizes the potential importance of new compounds and structures that display potent antifungal activity. The need for more and better antifungal agents has become more urgent because of the increasing incidence and severity of fungal infections associated with debilitating diseases, such as neoplasia, and with the administration of antibiotics, corticosteroids, and antineoplastic drugs (4–8). The few medicinal antifungal agents, such as nystatin, amphotericin B, and griseofulvin, are essentially devoid of antibacterial activity; and, conversely, antibacterial drugs are generally ineffective against pathogenic fungi. A new compound, methyl 5(or 4)-(3,3-dimethyl-1-triazeno)imidazole-4(or 5)-carboxylate (I, NSC-87982), with pronounced activity *in vitro* against a broad spectrum of both fungi and bacteria is reported here. This compound is structurally related to 5(or 4)-(3,3-dimethyl-1-triazeno)- and 5(or 4)-[3,3-bis(2-chloroethyl)-1-triazeno]imidazole-4(or 5)-carboxamide, both of which have antileukemic activity (9, 10).

The triazenoimidazole methyl ester (I) was prepared by diazotizing methyl 5(or 4)-aminoimidazole-4(or 5)-carboxylate with sodium nitrite and hydrochloric acid and then introducing a 25% aqueous solution of dimethylamine into the diazotization mixture. The product (I) was extracted with chloroform and, after evaporation of the chloroform, was twice dissolved in methanol and treated with activated carbon. Evaporation of methanol after the second decolorization gave I as a white solid that was homogeneous by thin-layer chromatography (silica gel, 9:1 chloroform–methanol), m.p. 163–165° dec. $\lambda_{max.}^{methanol}$ in mμ 233 (ε 10,500) and 328 (ε 16,700).

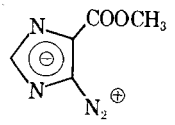
Anal.—Calcd. for $C_7H_{11}N_5O_2$: C, 42.64; H, 5.63; N, 35.52. Found: C, 42.56; H, 5.64; N, 35.23.

By extracting the diazotization reaction mixture with chloroform, the intermediate, methyl 5-diazoimidazole-4-carboxylate (II), may also be isolated as a crystalline solid, m.p. 95–96°, $\lambda_{max.}$ in mμ, 314 (ε 9400) and 270 (sh) in phosphate buffer (pH 7); $\bar{\nu}$ in cm.$^{-1}$: 2180 (N_2^+), 1700 (C=O).

Anal.—Calcd. for $C_5H_4N_4O_2$: C, 39.48; H, 2.65; N, 36.84. Found: C, 39.81; H, 2.98; N, 36.51.



I (NSC–87982)



II

Antimicrobial evaluation of NSC-87982 *in vitro* was performed by the paper disk–agar diffusion method previously described (11). NSC-87982 inhibits the growth of numerous strains of various microorganisms; the data shown in Table I illustrate its inhibitory action against representative yeasts, filamentous fungi, and Gram-positive and Gram-negative bacteria. Zones of inhibition produced by NSC-87982 in most of the fungal tests were comparable to, or greater than, those given by amphotericin B or by nystatin under similar conditions (12). It is less toxic to mice than certain 1,2,5-selenadiazoles that also have antifungal and antibacterial activity (13).

NSC-87982 significantly increased the lifespan of BDF_1 mice bearing lymphatic leukemia L1210. The average survival time of leukemic mice receiving chronic, intraperitoneal treatment (q.d. 1–30 days or to death) with NSC-87982 at 31 mg./Kg./day was approximately 160% of that of untreated leukemic mice. Inhibitory

TABLE I.—INHIBITION OF VARIOUS MICROORGANISMS BY NSC-87982[a]

Microorganisms	mcg./Disk of NSC-87982 1	10	100
Fungi			
Candida albicans ATCC 752	0	31	44
Hansenula wingei NRRL Y-2340	0	25	43
Saccharomyces carlsbergensis ATCC 9080	19	36	45
Rhodotorula sp. SRI 263	0	18	40
Venturia inequalis ATCC 11096	34	50	>60
Penicillium roqueforti ATCC 6987	0	16	45
Phycomyces nitens ATCC 9984	0	0	20
Aspergillus flavus ATCC 9643	0	0	28
Bacteria			
Pseudomonas aeruginosa ATCC 8709	tr	18	22
Salmonella typhimurium ATCC 7823	tr	17	21
Shigella sonnei ATCC 9290	0	13	19
Bacillus subtilis ATCC 10744	14	20	30
Staphylococcus aureus ATCC 6538	0	15	28
Mycobacterium phlei ATCC 354	23	32	44

[a] Numbers in table refer to the diameters in millimeters of the zones of inhibition of growth surrounding and including the disks. Diameter of disk is 12.7 mm. No inhibition is indicated by 0; tr = trace.

activity against fungi, neoplastic cells, and bacteria by a single compound is of special interest because of the danger of fatal fungal infections in cancer patients (4–6).

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Y. Fulmer Shealy
Charles A. Krauth
Robert F. Pittillo
Dale E. Hunt

Medicinal Chemistry Division
and Microbiology Division
Kettering-Meyer Laboratory
Southern Research Institute
Birmingham, Ala. 35205.

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Books

REVIEWS

Principles of Regulatory Drug Analysis. By Daniel Banes. Association of Official Analytical Chemists, P.O. Box 540, Benjamin Franklin Station, Washington, D. C. 20044, 1966. vi + 157 pp. 15.5 × 23.5 cm. Price $5.00 domestic, $5.25 foreign.

This book was written especially to point out the importance of chemical analysis in the effective enforcement of drug laws, regulations, and standards. The first chapter discusses the purpose of modern drug laws and the chemical evidence necessary to enforce them. The role of the U.S.P. and N.F. in establishing drug standards is also included. The remaining chapters are devoted to methods used in regulatory drug analysis using specific groups of drugs. The majority of the tests and assay procedures included are those appearing in the pertinent N.F. or U.S.P. or are those used by the FDA. Some discussion of methods which appear in "Official Methods of Analysis" of the A.O.A.C. is also given. The chemical principles upon which a particular test or assay is based are considered. Following each chapter, the author has included questions especially thought-provoking for students. The drug categories included are nonaddicting analgesics and antipyretics, barbiturates and related amides, sympathomimetics, antihistamines, estrogens, sulfonamides, and antibiotics.

Chemical Data Book. 2nd ed. Edited by G. H. Aylward and T. J. V. Findlay. John Wiley & Sons, Pty. Ltd., Sydney, Australia, 1966. viii + 88 pp. 15 × 24.5 cm. Paperbound. Price $2.95.

This small handbook serves as a source of data covering most of the chemical systems encountered by senior high school and first year college students. Included are properties of elements, inorganic, and organic compounds, some crystal forms, thermochemical data, and small tables of various constants and properties using only a few selected compounds.

NOTICES

Essentials of Biological Chemistry. 2nd Ed. By J. L. Fairley and G. L. Kilgour. Reinhold Publishing Corp., 430 Park Ave., New York, N. Y. 10022, 1966. xvii + 314 pp. 15.5 × 23.5 cm. Price $9.

Biochemistry: An Introduction to Dynamic Biology. By Ernest R. M. Kay. The Macmillan Co., 866 Third Ave., New York, N. Y. 10022, 1966. ix + 374 pp. 16 × 24 cm. Price $7.95.

Chemical Kinetics in Homogeneous Systems. By M. Ritchie. John Wiley & Sons, Inc., 605 Third Ave., New York, N. Y. 10016, 1966. viii + 115 pp. 14 × 21.5 cm. Price $2.95. Paperbound.

Outlines of Biochemistry. 2nd Ed. By Eric E. Conn and P. K. Stumpf. John Wiley & Sons, Inc., 605 Third Ave., New York, N. Y. 10016, 1966. x + 468 pp. 15.5 × 23.5 cm. Price $9.50.