Table I—Apparent Volumes of Distribution and Binding Parameters for Phenytoin in Healthy Volunteers, Nephrotic Patients, and Uremic Patients

Clinical Condition	V_β, liters/kg	f_p	f_T
Healthy volunteers (n = 6)[a]	0.3	0.10	0.22
Nephrotic patients (n = 6)[a]	0.6	0.19	0.19
Healthy volunteers (n = 4)[b]	0.64	0.12	0.11
Uremic patients (n = 4)[b]	1.40	0.26	0.11

[a] Apparent volumes of distribution and plasma binding data reported in Ref. 3. [b] Apparent volumes of distribution and plasma binding data reported in Ref. 4.

Table II—Apparent Volumes of Distribution and Binding Parameters for Warfarin in Rats

	V_β, liters/kg[a]	f_p[a]	f_T
High plasma binders (n = 6)	0.14	0.004	0.025
Low plasma binders (n = 7)	0.19	0.014	0.058

[a] Values were reported in Ref. 5.

plasma binding data were obtained from the literature (3–5). Total body water was assumed to be 599 ml/kg in humans and 660 ml/kg in rats. Plasma volume was assumed to be 46 ml/kg in humans and 42 ml/kg in rats.

The findings with phenytoin are summarized in Table I. Both types of renal disease resulted in a twofold increase in both the fraction of free drug in the plasma and the apparent volume of distribution compared to those observed in healthy volunteers. Consequently, in neither case was the apparent tissue binding affected. The reduced plasma binding observed in nephrotic patients was essentially secondary to hypoalbuminemia (3), whereas that observed in uremic patients was presumably due to changes in the molecular structure of albumin or to the accumulation of an avidly bound endogenous inhibitor that acts like a displacing agent (12).

In view of the unaltered tissue binding of phenytoin in nephrotic patients, either hypoalbuminemia in the extravascular space is less pronounced than that observed in the vascular space or molecules other than albumin are principally responsible for binding phenytoin in the tissues. In uremic patients, phenytoin binding sites in the tissue are less susceptible to disease-related changes than phenytoin binding sites in the plasma.

As shown in Table II, differences in plasma protein binding of warfarin in rats were paralleled by qualitatively similar differences in tissue binding. Rats that showed relatively high plasma binding of warfarin also showed relatively high tissue binding of the drug. Consistent with this finding is the observation that the fraction of warfarin unbound to liver homogenates correlated strongly with the fraction of warfarin unbound to serum in the same animals (5). Perhaps a similar factor is responsible for the intersubject variability of warfarin binding in both plasma and tissue of rats.

The examples presented illustrate the utility, at least in a conceptual sense, of considering drug distribution in a physiological framework (10). Estimations of apparent tissue binding can provide guidelines for more extensive distribution studies.

(1) B. Alexanderson, *Eur. J. Clin. Pharmacol.*, **6**, 44 (1973).

(2) M. M. Ghoneim and H. Pandya, *Anesthesiology*, **42**, 545 (1975).

(3) R. Gugler, D. W. Shoeman, D. H. Huffman, J. B. Cohlmia, and D. L. Azarnoff, *J. Clin. Invest.*, **55**, 1182 (1975).

(4) I. Odar-Cederlöf and O. Borga, *Eur. J. Clin. Pharmacol.*, **7**, 31 (1974).

(5) A. Yacobi and G. Levy, *J. Pharm. Sci.*, **64**, 1660 (1975).

(6) W. J. Jusko and M. Weintraub, *Clin. Pharmacol. Ther.*, **16**, 449 (1974).

(7) R. H. Reuning, R. A. Sams, and R. E. Notari, *J. Clin. Pharmacol.*, **13**, 127 (1973).

(8) G. R. Wilkinson and D. G. Shand, *Clin. Pharmacol. Ther.*, **18**, 377 (1975).

(9) G. R. Wilkinson and S. Schenker, *Biochem. Pharmacol.*, **25**, 2675 (1976).

(10) J. R. Gillette, *Ann. N.Y. Acad. Sci.*, **179**, 43 (1971).

(11) M. Gibaldi, R. Nagashima, and G. Levy, *J. Pharm. Sci.*, **58**, 193 (1969).

(12) D. S. Campion, *Toxicol. Appl. Pharmacol.*, **25**, 391 (1973).

Milo Gibaldi [x]
Patrick J. McNamara

Department of Pharmaceutics
School of Pharmacy
State University of New York at Buffalo
Buffalo, NY 14260

Received March 28, 1977.

Accepted for publication May 5, 1977.

Supported in part by Grant GM-20852 from the National Institutes of Health.

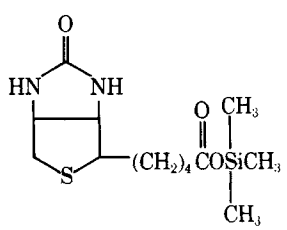
[x] To whom inquiries should be directed.

Characterization of Biotin Trimethylsilyl Derivative

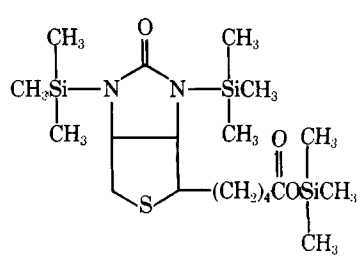
Keyphrases □ Biotin trimethylsilyl derivative—structure determined □ Trimethylsilyl derivative of biotin—structure determined □ GLC–mass spectrometry—analysis, trimethylsilyl derivatives of biotin, structure determined □ Vitamins—biotin, structure of trimethylsilyl derivative determined

To the Editor:

Analytical methods are available for the determination of biotin (1, 2). In view of the need for a stability-indicating assay procedure, the GLC method (2) was investigated. The purpose of this communication is to report that the silyl derivative actually prepared according to this procedure is not the silyl ester reported (I) but the trisilyl de-



I



II

rivative (II) with the proposed structure indicated. It seems appropriate to correct the record now since I was again proposed as the structure for silylated biotin in a recent review (3).

The possibility that the proposed structure for the silyl ester (I) might be incomplete appeared when we prepared the methyl ester of biotin using diazomethane. Although this methyl ester could be chromatographed, the compound had a much longer retention time compared with the silylated compound and produced a peak that tailed badly. This large difference in chromatographic behavior is not expected for two compounds that are supposed to be simple esters. It also was observed that when the silylating agent [bis(trimethylsilyl)trifluoroacetamide] (III) was added to the previously formed methyl ester, the late eluting broad peak disappeared and a sharp, early eluting peak was obtained. These observations led to a more definitive study of the derivatives formed.

The fully silylated derivatives can be formed by reacting 5 mg of biotin with 1 ml of a 1:1 (v/v) solution of III and pyridine. After heating at 60° for 15 min, the sample is ready for analysis. The partially silylated derivative can be formed by first reacting 5 mg of biotin in ether with excess diazomethane until the yellow color persists. After 10 min, the methyl ester solution is evaporated to dryness and then silylated as described.

These samples were then analyzed[1] on a gas chromatograph–mass spectrometer[2] using a 1.9-m 3% OV-17 on 100–120-mesh Gas Chrom Q[3] column. The mass spectrum of the fully silylated derivative produced molecular ion 460 together with a base peak at *m/e* 73, supporting Structure II. The mass spectrum of the methylated biotin trimethylsilyl derivative gave *m/e* 402, again with a base peak at *m/e* 73, supporting the biotin methyl ester *N,N*-trimethylsilyl structure.

(1) C. Plinton, F. P. Mahn, M. Hawrylyshyn, V. S. Venturella, and B. Z. Senkowski, *J. Pharm. Sci.*, **58**, 875 (1969).
(2) V. Viswanathan, F. P. Mahn, V. S. Venturella, and B. Z. Senkowski, *ibid.*, **59**, 400 (1970).
(3) S. Ahuja, *ibid.*, **65**, 163 (1976).

Raymond N. Johnson [x]
George R. Boyden
Analytical Research and Development Laboratory
Ayerst Laboratories, Inc.
Rouses Point, NY 12979

Received February 24, 1977.
Accepted for publication May 24, 1977.
[x] To whom inquiries should be directed.

[1] Courtesy of Dr. G. Schilling, Analytical Research, Ayerst Laboratories, Montreal, Quebec, Canada.
[2] LKB 9000.
[3] Applied Science Laboratories, State College, Pa.

BOOKS

REVIEWS

Annual Reports in Medicinal Chemistry. Vol. 11. Edited by FRANK H. CLARKE *et al.* Academic, 111 5th Ave., New York, NY 10003, 1976. 339 pp. 17 × 25.5 cm. Price $16.50.

The new editor-in-chief of this series, Dr. Frank Clarke, is to be congratulated on maintaining the quality of content, presentation, and timeliness found in earlier volumes. The 32 chapters of this volume are grouped into six principal sections. Six section editors (three of them new) have the responsibility for the contributions in their areas.

Most chapters survey the literature of 1975 on new compounds reported to have pharmacologic activity. A few chapters cover the years since a report last appeared in this series. Some of the most interesting chapters present a mini-review of some biological process or disease state and point the way to potential new approaches for drug therapy. A third type of chapter deals with general methods of possible utility in the synthesis, design, or testing of drugs.

The section on CNS Agents has three chapters that update previous coverage of antidepressants, antipsychotic agents, antianxiety agents, anticonvulsants, sedative–hypnotics, analgesics (including endogenous peptides), and narcotic antagonists. In addition, there are two excellent chapters on the currently "hot" areas of opiate receptors and biological factors in psychiatric disorders. The Pharmacodynamic Agents section covers pulmonary and antiallergy drugs, antihypertensives, diuretics, and the structure–activity relationships of prostaglandins. The Chemotherapeutic Agents section has chapters on antibiotics and antifungal, antiviral, antiparasitic, and antineoplastic agents.

The section on Metabolic Diseases and Endocrine Function has chapters devoted to immunosuppressive and immunostimulatory agents, steroids, peptide hormones, diabetes, disorders of lipid metabolism, drug metabolism, and antiobesity agents. The Topics in Biology section features particularly pertinent chapters on membrane regulators, active transport across membranes, the antimetabolite concept, comparative toxicology, and chronopharmacology. The last section on Topics in Chemistry has the usual chapter on synthetic reactions of interest plus chapters concerning the synthesis of β-lactam antibiotics, synthesis of cyclic adenosine monophosphate and cyclic guanidine monophosphate derivatives, syntheses employing polymeric reagents, quantitative drug design, and use of NMR for drug binding studies.

As expected with any work by 46 different authors, the writing style is far from uniform. However, the readability of this volume is generally quite good. Every medicinal chemist and most pharmacologists would surely find several chapters of interest.

Reviewed by James F. Stubbins
Virginia Commonwealth University
Richmond, VA 23298

Chinese Herbs: Their Botany, Chemistry, and Pharmacodynamics. By JOHN D. KEYS. Tuttle, Rutland, VT 05701, 1976. 338 pp. 15 × 22 cm. Price $15.00.

This book is a compendium of monographs of Chinese medicinal plants arranged by botanical classification. Each monograph includes an illustration and Chinese characters as well as a botanical, chemical, and pharmacological description. There are also supplementary sections on mineral- and animal-derived drugs, a collection of Chinese prescriptions, and a table of toxic herbs.

This book is a good compilation of the medicinal plants of China, done mainly through translation of Chinese works, some as old as the 6th century. The botanical description seems quite complete, while the chemical and pharmacodynamic sections appear too outdated and su-