trometer[3]. The mass spectrum obtained (Fig. 1) contained additional mass peaks, one unit greater than the normal ^{12}C-containing peaks, in an abundance indicating massive carbon-13 incorporation into the constituents of the white fly tissue.

(1) J. J. Katz, R. A. Uphaus, H. L. Crespi, and M. I. Blake, in "Isotopes and Chemical Principles," P. A. Rock, Ed., ACS Symposium Series, American Chemical Society, Washington, D.C., 1975.

(2) M. I. Blake, H. L. Crespi, and J. J. Katz, *J. Pharm. Sci.*, **64**, 367(1975).

(3) R. A. Uphaus, "Proceedings of the First International Conference on Stable Isotopes in Chemistry, Biology and Medicine, May 9–11, 1973," Argonne National Laboratory, Argonne, Ill., 1973.

(4) R. A. Uphaus, M. I. Blake, and J. J. Katz, *Can. J. Bot.*, **56**, 352(1975).

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Received December 1, 1975.

Accepted for publication March 4, 1976.

Presented in part at the Pharmacognosy and Natural Products Section, APhA Academy of Pharmaceutical Sciences, Chicago meeting, August 1974.

Performed under the auspices of the Atomic Energy Commission with the aid of a grant from the American Medical Association Tobacco Research Fund.

Supported in part by AMA Tobacco Contract P-7146-C.

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[3] The authors are indebted to Dr. Martin Studier, Argonne National Laboratory, for the mass spectral data.

Biased Bioavailability Estimates

Keyphrases □ Bioavailability—griseofulvin, use of fractional urinary excretion data questioned □ Griseofulvin—bioavailability, use of fractional urinary excretion data questioned □ Urinary excretion data, fractional—value in griseofulvin bioavailability studies questioned □ Antifungal agents—griseofulvin bioavailability, use of fractional urinary excretion data questioned

To the Editor:

In a recent communication (1), followed by publication of the detailed data (2), Bates and Sequeira proposed that 24-hr urinary excretion data of total (free and glucuronide conjugated) 6-desmethylgriseofulvin, the main metabolite of griseofulvin, be used to determine the bioavailability of the parent drug. However, I wish to point out that this general practice (1, 3, 4) of using fractional urinary drug excretion data and/or areas under the plasma level–time curve without proper pharmacokinetic justification, even with apparent correlations with total urinary excretion and total area under the plasma level–time curve data, is dangerous.

Table I—Relative Bioavailability of Formulations I–IV of Griseofulvin Calculated from the Ratios of the Total Urinary Metabolite at 24 and 96 hr[a]

Formulation[b]	24 hr	96 hr
I:IV	0.32	0.43
II:IV	0.41	0.53
III:IV	0.52	0.60
I:III	0.61	0.71
II:III	0.78	0.87
I:II	0.79	0.81

[a] Data are from Fig. 1 and Table II of Ref. 2. The differences among the relative bioavailabilities calculated from the urinary excretion data of total metabolite at 24 and 96 hr are statistically significant; two-tailed paired *t*-test: t (0.002) = 5.89, t calc = 5.97. [b] I = aqueous suspension, II = commercial Tablet A, III = commercial Tablet B, and IV = corn oil-in-water emulsion.

Bates and Sequeira (1) stated that, while the plasma levels of the entirely metabolized griseofulvin are usually low after oral administration of different formulations, the measurement of 24-hr urinary excretion of the main metabolite, 6-desmethylgriseofulvin, could be more reliable and more convenient since the use of a 72–96-hr urinary collection period "increases the chances for a lack of compliance to the experimental protocol on the part of subjects . . . ." These investigators reported that a correlation existed between the dose percentages of total 6-desmethylgriseofulvin excreted within 24 and 96 hr after administration of various formulations of griseofulvin and concluded that: "These correlations . . . provide the basis for possible utilization of 24-hr cumulative total 6-desmethylgriseofulvin excretion data as an index of griseofulvin bioavailability in humans."

However, an accurate assessment of the relative bioavailability of a given griseofulvin formulation from metabolite cumulative recovery excretion data measured over a limited 24-hr time interval is only possible if the absorption process of griseofulvin is complete much sooner than 24 hr after drug administration. However, with some oral formulations of griseofulvin, the absorption process occurs for 30–40 hr and even up to 80 hr (5). The bioavailability of such formulations would be underestimated by the use of 24-hr cumulative metabolite excretion data, since the total fraction of the dose administered that will eventually be absorbed is not totally absorbed at 24 hr after administration.

Urinary excretion data of the metabolite (free and glucuronide conjugated) after oral administration permit only the calculation of relative bioavailabilities of different formulations given by the same route of administration. Since griseofulvin is entirely metabolized (6) [most likely in the liver (7)], a first-pass effect, although not large (8–19% of the dose absorbed), must be anticipated (8). The linearity of an existing first-pass effect needs to be experimentally challenged. The fact that ultimately the amounts of free 6-desmethylgriseofulvin excreted in the urine are constant fractions of dosages of formulations with widely differing absorption characteristics does not negate a possible nonlinearity of griseofulvin metabolism (2). Parent drug data ob-

tained in intravenous and oral studies are indispensable for this purpose. Differences in the ratio of free to glucuronide-conjugated 6-desmethylgriseofulvin observed after different oral formulations (2) appear to be indicative of nonlinear kinetics for these metabolites.

The fact that highly significant statistical correlations apparently exist between 24- and 96-hr urinary metabolite excretions of a given formulation does not prove that 24-hr urinary metabolite excretion data are accurate and adequate for bioavailability assessments of griseofulvin. The existence of these correlations indicates only that a formulation with higher bioavailability will tend to give higher urinary metabolite levels at 24 and 96 hr than a formulation with lower bioavailability at these times.

Notwithstanding an apparent correlation, the bias in estimating relative bioavailabilities of different formulations using 24- rather than 96-hr urinary excretion total metabolite data can be demonstrated with the data reported by Bates and Sequeira (2). They investigated the cumulative urinary excretion of total 6-desmethylgriseofulvin after the administration of four different griseofulvin formulations to five individuals. On the assumption of first-order kinetics, the relative bioavailability of the four different griseofulvin formulations can be calculated from the ratios of the mean amounts of total metabolite excreted in urine at different times (Table I).

It is evident that the use of 24-hr urinary excretion data of total metabolite leads consistently to underestimations of derived relative bioavailabilities. The underestimation increases with the smaller bioavailabilities, consistent with the premise that delayed absorption is the primary reason for low bioavailability. Thus, underestimation will be greatest when the relative bioavailability of poorly absorbed formulations is studied[1].

The standard error of the mean, expressed as percent of the mean, was consistently larger for the total amount of urinary metabolite at 24 hr. At 24 hr, the values were: I, 11.5%; II, 18.3%; III, 15.6%; and IV, 4.6%. At 96 hr, they were: I, 5.4%; II, 12.2%; III, 10.6%; and IV, 4.2%. Thus, routine assessments of bioavailabilities of different griseofulvin formulations should not be based on 24-hr urinary excretion data of total 6-desmethylgriseofulvin even if first-order kinetics exist.

I feel strongly that this dangerous procedure of using fractional urinary excretion data and/or areas (1, 3, 4) for the assessment of bioavailability must be clearly recognized.

(1) T. R. Bates and J. A. Sequeira, *J. Pharm. Sci.*, **64**, 709(1975)
(2) *Ibid.*, **64**, 793(1975).
(3) D. J. Greenblatt, D. W. Duhme, J. Koch-Weser, and T. W. Smith, *Clin. Pharmacol. Ther.*, **16**, 813(1974).
(4) E. Steiness, V. Christensen, and H. Johansen, *ibid.*, **14**, 949(1973).
(5) W. L. Chiou and S. Riegelman, *J. Pharm. Sci.*, **60**, 1376 (1971).
(6) C.-C. Lin, J. Magat, R. Chang, J. McGlotten, and S. Simchowicz, *J. Pharmacol. Exp. Ther.*, **187**, 415(1973).
(7) M. Rowland, S. Riegelman, and W. L. Epstein, *J. Pharm. Sci.*, **57**, 984(1968).
(8) P. A. Harris and S. Riegelman, *ibid.*, **58**, 93(1969).

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Received July 14, 1975.
Accepted for publication March 30, 1976.

[1] The rank order for the bioavailability of products in Table I is the same at 24 as it is at 96 hr (IV > III > II > I).

BOOKS

REVIEWS

Aliphatic Chemistry, Volume 3, Specialist Periodical Report. Senior Reporter, A. McKILLOP. The Chemical Society, Burlington House, London WIV OBN, England, 1975. xii + 409 pp. 14.5 × 22 cm. Price £13.50.

The first volume of "Aliphatic Chemistry" was published as Part I of a three-part Specialist Periodical Report on aliphatic, alicyclic, and saturated heterocyclic chemistry. Subsequently, these areas have been reviewed in separate Specialist Periodical Reports. This Specialist Report is the third volume of the series, dealing with aliphatic chemistry and, except for one chapter, reviews the literature published during 1973. Chapter 1, by R. S. Atkinson, is devoted to acetylenes, alkanes, allenes, and alkenes; Chapter 2, by E. W. Colvin, surveys the literature on other functional groups (carboxylic acids, esters, lactones, anhydrides, amides, nitriles, aldehydes and ketones, alcohols, amines, alkyl halides, ethers, and sulfur compounds). These chapters are similar in format and style to analogous chapters by the same authors in the two preceding volumes.

This volume, like Volume 2, contains chapters that summarize progress in the area of naturally occurring polyolefinic and polyacetylenic compounds (Chapter 3) and in the chemistry of the prostaglandins (Chapter 4). Both chapters are authored by G. Pattenden, as were the corresponding chapters in Volume 2. The literature on fatty acids and related compounds was reviewed in Volume 1 but not in Volume 2. Developments in this area during both 1972 and 1973, therefore, comprise Chapter 5 by F. D. Gunstone. The specialized areas of Chapters 3–5 are exemplified by sections dealing with polyolefinic antibiotics and other microbial metabolites, insect pheromones, acetylenes and olefins of marine or plant origin, prostaglandin syntheses, the multifaceted aspects of studies of fatty acids, and others.

A cumulative set of volumes may serve as summaries of the literature for the specialist engaged in research in one of these areas; but, perhaps more importantly, these annual surveys will provide a means by which others may be introduced to, or become familiar with, developments in a specific area. The value of surveys of the annual literature devoted to specific areas, such as those of Chapters 3–5, is readily recognized. Reviews of the more general topics of the first two chapters are equally valuable, since much effort would be required for the individual to extract this type of information from the periodical or abstract literature. In giving comprehensive coverage of the significant literature, the authors maintain the high standards of the preceding volumes.

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