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▲ To whom inquiries should be directed.

COMMUNICATIONS

Inhibited Dissolution of Drug Crystals by Certified Water-Soluble Dyes: *In Vivo* Effect

Keyphrases □ Sulfathiazole dissolution—*in vivo* inhibition by water-soluble dye, man □ Dissolution, sulfathiazole—*in vivo* inhibition by water-soluble dye, man □ Dyes, water soluble—*in vivo* inhibition of sulfathiazole dissolution, man

Sir:

In previous articles, Piccolo and Tawashi (1–4) established the inhibitory effect of low concentrations of various water-soluble dyes on the dissolution rate of crystalline drugs. The effect of the degree of undersaturation, the effect of the nature of the dye, and the dependence of the dissolution rate on the dye concentration in powder systems were confirmed. The results obtained were discussed in the light of preferential adsorption of the dye molecules on the primary dissolution sources of the crystal surface. Further experiments were conducted to determine the influence of FD&C Blue No. 1 on the solubilizing effect by 0.04 *M* sodium cholate, using the sulfathiazole single crystal as a model substance. The data obtained showed that a concentration of only 5 mcg./ml. of the dye inhibited the dissolution rate to a value very close to that in distilled water (3). Since dissolution rate is often a rate-limiting step in the absorption of drugs with low aqueous solubility, and since micellar solubilization by bile salts plays an important role in the intestinal absorption of these drugs, the results obtained with certified water-soluble dyes are extremely interesting in relation to the drug absorption process. The present study was undertaken to examine the effect of FD&C Blue No. 1 on the absorption rate of sulfathiazole in man. Sulfathiazole Form I and FD&C Blue No. 1 were selected because they were tested *in vitro* previously (1, 3).

Three healthy young adult male human subjects were used in this study. One gram of sulfathiazole Form I crystals, 40 mesh USP, was suspended in 200 ml. of water and administered to each subject. Blood samples (0.2 ml., by finger puncture) were withdrawn at 0, 0.5, 1, 1.5, 2, 2.5, 3, 4, and 5 hr. following administration. After an interval of 1 week, a fresh suspension of 1 g. of sulfathiazole crystals in 200 ml. of water containing 10 mcg./ml. of FD&C Blue No. 1 (total quantity of dye is 2 mg.) was prepared and administered. Administration was in the morning to subjects in the fasting state, and no food was ingested until withdrawal of the last blood sample (5 hr.).

The blood samples (0.2 ml.) were diluted with 3 ml. of distilled water and precipitated with 0.8 ml. of 20% trichloroacetic acid. After centrifugation, the concentrations of free sulfathiazole were determined by the method of Bratton and Marshall (5), modified to use 2 ml. of the clear supernate (equal to 0.05 ml. of blood), 0.2 ml. of 1% sodium nitrite, 0.2 ml. of 0.5% ammonium sulfamate, and 1 ml. of 0.05% *N*-(1-naphthyl)ethyl-

Table I—Free Sulfathiazole Blood Concentrations for Three Subjects after Ingestion of 1 g. Oral Doses in the Absence and in the Presence of FD&C Blue No. 1

	Blood Concentrations, mg./100 ml.							
	Subject A		Subject B		Subject C		Mean Values	
Hours	Without Dye	With Dye	Without Dye	With Dye	Without Dye	With Dye	Without Dye	With Dye
0.5	0.285	0.116	0.381	0.174	0.254	0.000	0.307	0.097
1	0.793	0.445	0.841	0.237	0.444	0.175	0.693	0.286
1.5	0.888	0.889	1.412	0.365	0.746	0.635	1.015	0.630
2	1.349	1.064	1.412	0.936	1.047	0.746	1.236	0.915
2.5	1.793	0.873	1.190	0.984	1.206	0.826	1.396	0.894
3	1.555	1.064	1.174	1.158	1.270	0.937	1.333	1.053
4	1.714	1.032	1.365	1.444	1.190	1.588	1.423	1.355
5	2.418	1.127	2.016	1.571	1.254	1.635	1.896	1.444

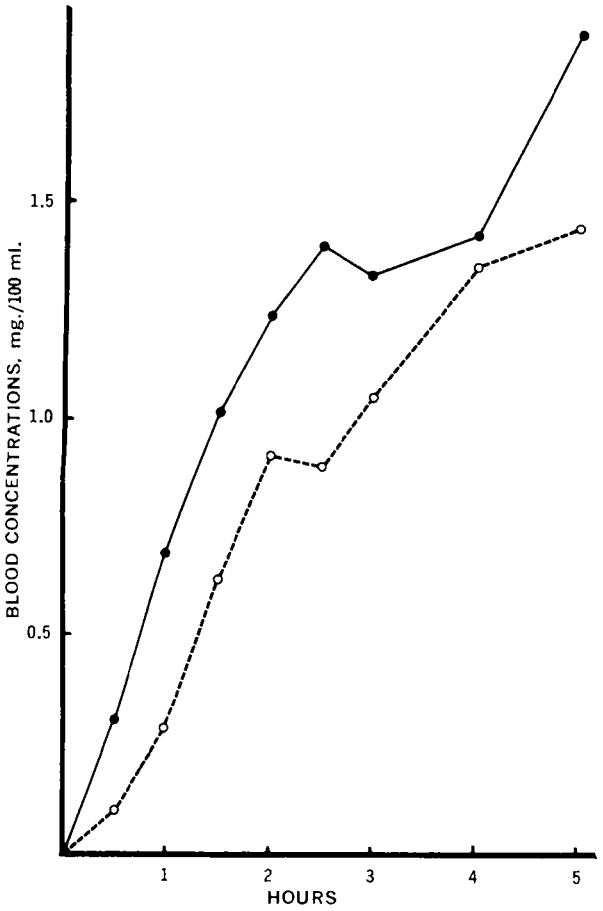

Figure 1—*Blood concentrations of free sulfathiazole following oral administration of 1 g. sulfathiazole crystals. Key: ○, in presence of FD&C Blue No. 1; and ●, in absence of FD&C Blue No. 1.*

enediamine dihydrochloride (6). Spectrophotometric measurements were carried out on the final colored solutions at a wavelength of 543 nm., and comparisons were made with an appropriate standard. Blanks prepared from control blood samples, from each subject, were used for appropriate corrections.

Table I shows blood concentrations of sulfathiazole obtained following administration in the absence and in the presence of Blue No. 1. From these results, it is apparent that, in the first 3 hr., the sulfathiazole concentrations in the presence of the dye were lower than those in the absence of the dye for all three subjects. Figure 1 presents the blood concentration curves as a function of time obtained from the mean values presented in Table I. The data obtained in this study are in excellent accord with the *in vitro* dissolution rate data (1, 2). This agreement suggests that such a small concentration of FD&C Blue No. 1 could delay significantly the absorption of sulfathiazole through dissolution inhibition.

It should be stressed, however, that the limited data presented in this report on sulfathiazole crystals and Blue No. 1 cannot exclude the possibility that other dyes can exert similar effects on the absorption of crystalline drugs with poor solubility. More extensive studies are desirable with regard to absorption kinetics in the presence of certified dyes.

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R. TAWASHI▲
J. PICCOLO
Faculty of Pharmacy
University of Montreal
Montreal, Canada 101

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▲ To whom inquiries should be directed.

Fagaronine, a New Tumor Inhibitor Isolated from *Fagara zanthoxyloides* Lam. (Rutaceae)

Keyphrases □ *Fagara zanthoxyloides* Lam.—isolation, identification of fagaronine, antitumor activity □ Fagaronine—isolation, identification from *Fagara zanthoxyloides* Lam., antitumor activity □ Antitumor activity—fagaronine, isolated from *Fagara zanthoxyloides* □ Medicinal plants—isolation, identification of fagaronine from *Fagara zanthoxyloides* Lam., antitumor activity

Sir:

During a phytochemical investigation of *Fagara zanthoxyloides*[1], we isolated a new alkaloid for which we have assigned the trivial name fagaronine. Fagaronine has been shown to elicit a high order of activity against the P-388 leukemia in mice, giving prolongations of life on the order of 265, 210, and 190% T/C at doses of 100, 50, and 25 mg./kg., respectively. A compound is active if it exhibits a T/C of ≧125% (1)[2]. Several leukemic mice treated with fagaronine were considered as "cures."

Fagaronine crystallized as the chloride (from a mixture of ethyl acetate–methanol) as bright-yellow needles, exhibiting m.p. 202° followed by solidification and melting again at 255°[3]. A UV absorption spectrum of λ_{max} 233 (log ϵ 4.29), 272 (4.55), 305 (4.44) (sh), and 328 nm. (4.44) indicated that fagaronine belonged to the benzophenanthridine class of alkaloids; in particular, this spectrum resembled that of nitidine (2). The UV spectrum of fagaronine in 0.01 *N* HCl showed no change; however, in 0.01 *N* NaOH, a bathochromic shift to λ_{max} 346 nm. (log ϵ 4.31) was observed, suggest-

[1] The plant material was collected in Ghana by Mr. O. B. Dokosi, University of Ghana. A voucher specimen (SP-280) was identified by Mr. Dokosi as *F. zanthoxyloides* Lam. (Rutaceae) and is deposited in the Herbarium of the Department of Pharmacognosy and Pharmacology, University of Illinois at the Medical Center, Chicago, Ill. The roots were the parts used.

[2] The antitumor tests were performed through the Drug Research and Development Branch, National Cancer Institute, Bethesda, Md.

[3] Details of the isolation of fagaronine will be published.