Bioavailability of Dyphylline and Dyphylline–Guaifenesin Tablets in Humans

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Abstract □ A six-way-crossover bioavailability study was conducted with twelve healthy male volunteers to evaluate the relative bioavailability of three tablet formulations containing dyphylline and three tablet formulations containing dyphylline–guaifenesin. Each subject was administered two tablets of each product with ≥3 d separating each dose. Blood samples were obtained just prior to each dose and at 0.25, 0.5, 0.75, 1.0, 1.5, 2.0, 3.0, 4.0, 6.0, 8.0, and 10.0 h following each dose. An HPLC method was used to assay dyphylline in the serum. The mean t_{max} ranged from 0.6 to 1.0 h for the six products. The mean values for C_{max} differed by 29%, and the AUC values differed by <8%. It was noted that the dyphylline–guaifenesin products exhibited a lower bioavailability than the products which only contained dyphylline. It was concluded that the three combination products were bioequivalent, as were the three dyphylline products.

Dyphylline, a xanthine derivative possessing peripheral vasodilator and bronchodilator action, has been reported to have fewer side effects than equal doses of theophylline; however, its bronchodilator potency is significantly less.[1] Further, the elimination half-life of ~2 h is significantly shorter than that reported for theophylline, and dyphylline is primarily eliminated unchanged in the urine.[2,3] Several studies have evaluated the absorption of dyphylline after oral administration as solutions,[4,5] immediate release,[2-6] or controlled-release dosage forms.[4,6] However, no studies have been reported comparing the bioavailability of marketed tablets of dyphylline in combination with guaifenesin. The objective of this study was to evaluate the bioavailability of such marketed products in healthy human volunteers.

Experimental Section

Dosage Forms—Six tablet formulations containing 200 mg of dyphylline were obtained through commercial sources. Three of the formulations contained only dyphylline [product 1 (Lufyllin-200, Wallace Labs, lot 53N); product 3 (Dilor-200, Savage Labs, lot 9804); product 5 (Neothylline-200, Lemmon, lot 8486)] and the other three also contained 200 mg of guaifenesin[2] [product 2 (Lufyllin-GG, Wallace Labs, lot 82N); product 4 (Dilor-G, Savage Labs, lot 2819); product 6 (Neothylline-GG, Lemmon, lot 8530)].

Clinical Protocol—Twelve healthy, nonsmoking male volunteers were selected on the basis of a medical history, blood chemistry, hematology, and urinalysis. They ranged in age from 21 to 32 years and weighed 66–93 kg. The subjects were instructed to refrain from all medications for the week prior to the study and during the study period. No alcohol or xanthine containing foods or beverages were allowed for 24 h prior to each treatment period and for 10 h after receiving each dose. A randomized, incomplete block, factorial design was employed, with each subject receiving two tablets of each of the six formulations. The study was conducted over a 3-week period, with ≥3 d between each dose. The doses were administered with 240 mL of water in the morning after an overnight fast. No food or beverage other than water was permitted for 4 h after dosing, at which time a standard meal was provided.

Blood samples (8 mL) were collected from the subjects just prior to each dose and then at 0.25, 0.5, 0.75, 1.0, 1.5, 2.0, 3.0, 4.0, 6.0, 8.0, and 10.0 h following each dose. The samples remained at ambient temperature for 1 h, were centrifuged, and the serum fraction stored frozen until analysis.

Analytical Methods—Dyphylline serum concentrations were determined in duplicate using an HPLC procedure based on a method reported by Gisclon et al.[7] Standard curves were prepared daily in duplicate by combining 1 mL serum, 0.5 mL of aqueous dyphylline standard solution (0.25–8.0 μg/mL), 0.5 mL of β-hydroxyethyltheophylline internal standard (3 μg/mL), and 0.5 mL of 0.4 M NaOH in a 20-mL screw-cap centrifuge tube. After the addition of 10 mL of isopropyl alcohol:chloroform (20:80, v/v) the mixture was shaken vigorously on a platform shaker for 20 min and centrifuged at 3000 rpm at −10°C for 25 min. The aqueous layer was discarded and the organic phase was transferred to a silanized conical tube and evaporated with nitrogen at 40°c. The dried residue was reconstituted with mobile phase and 5 μL was injected into the HPLC system (Hewlett-Packard 1081B HPLC and 21MX Computer System; Waters Associates 440 Detector and μ-Bondapak C_{18}, 30-cm column.). No interference from guaifenesin was noted in chromatograms obtained from samples containing dyphylline and internal standard. The standard curves, plotted as peak area ratio (dyphylline:internal standard), exhibited excellent linearity ($r \geq 0.999$). Five standard curves prepared with duplicate samples over a 5-d period resulted in RSD values of 8.8% for the 0.25-μg/mL lowest standard and 1.2% for the 8.0-μg/mL highest standard.

Data Analysis—The individual time of maximum serum concentration (t_{max}) and maximum serum concentration (C_{max}) were obtained directly from serum concentration–time profiles. The area under the serum concentration–time curve (AUC_{0-10h}) was determined using the trapezoidal rule. The terminal disposition rate constant (k), half-life ($t_{1/2}$), area under the serum concentration time–curve to infinity ($AUC_{0-\infty}$), and the mean residence time (MRT) were estimated using standard model-independent techniques.[8]

Mean values for individual blood sampling times and each of the parameters cited above were statistically analyzed using analysis of variance and the Newman–Keuls a posteriori test. A power analysis[9] was employed to estimate the potential for statistical errors based on $\alpha = 0.05$ and $\beta = 0.2$.

Dissolution Testing—Six tablet formulations from the lots used in the bioavailability studies were tested in vitro using the USP Method II. Since, at the time of testing, dyphylline tablets were not official in the USP, the evaluation employed 900 mL of distilled water at 37°C, with a stirring rate of 50 rpm. Each tablet was tested in triplicate, with 5 mL of medium removed at 5, 10, 15, 30, 45, 60, 90, and 120 min. Each volume removed was replaced with distilled water, and the aliquots were diluted

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Table I—Dyphylline Serum Concentrations (μg/mL) at Each Sampling Time[a]

Product No.	0.25 h	0.5 h	0.75 h	1 h	1.5 h	2 h	3 h	4 h	6 h	8 h	10 h
1	3.12	7.45	6.91	5.66	4.36	3.47	2.57	1.82	0.89	0.46	0.26
	(59)	(41)	(28)	(25)	(25)	(15)	(20)	(21)	(23)	(27)	(38)
2	0.77	3.28	4.65	4.92	4.64	3.81	2.78	2.00	1.02	0.48	0.28
	(80)	(66)	(39)	(33)	(13)	(14)	(14)	(14)	(26)	(24)	(29)
3	2.71	6.97	6.47	5.62	4.45	3.72	2.63	1.84	0.90	0.46	0.26
	(83)	(38)	(26)	(18)	(15)	(15)	(18)	(23)	(24)	(36)	(44)
4	1.36	5.50	6.68	5.49	4.27	3.53	2.54	1.83	0.87	0.39	0.24
	(69)	(49)	(25)	(16)	(11)	(15)	(19)	(20)	(30)	(37)	(50)
5	2.34	7.08	6.53	5.69	4.59	3.70	2.65	1.91	0.91	0.50	0.26
	(87)	(47)	(38)	(21)	(18)	(16)	(18)	(25)	(32)	(33)	(44)
6	1.44	4.68	5.49	5.16	4.22	3.63	2.68	2.00	0.98	0.50	0.27
	(86)	(52)	(22)	(24)	(21)	(22)	(27)	(34)	(40)	(37)	(41)
Percent Difference[b]	75.3	56.0	32.7	13.5	9.1	8.9	8.6	9.0	14.7	22.0	14.3

[a] Each value represents the mean of 12 subjects. The RSD values are given in parentheses (SD × 100/mean). [b] (Highest − Lowest)(100)/(Highest).

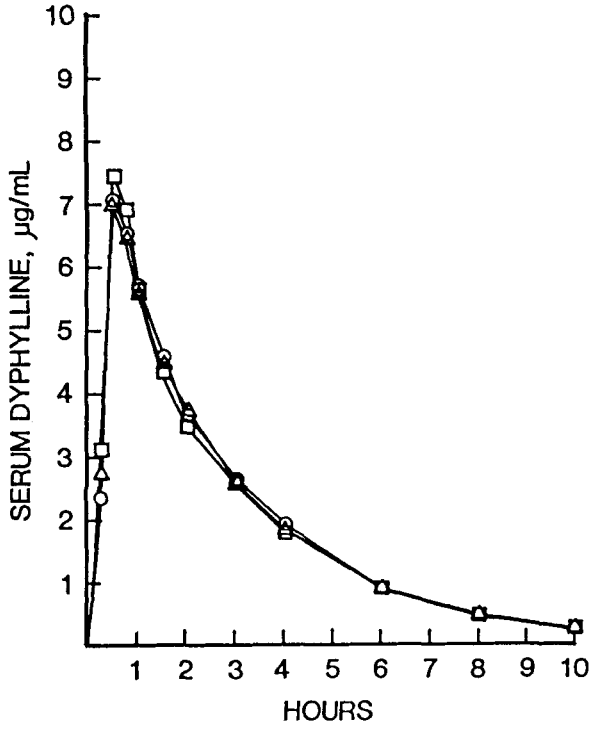

Figure 1—*Mean (n = 12) serum dyphylline concentrations after single doses (two tablets) containing 200 mg of dyphylline. Key: (□) product 1; (△) product 3; (○) product 5.*

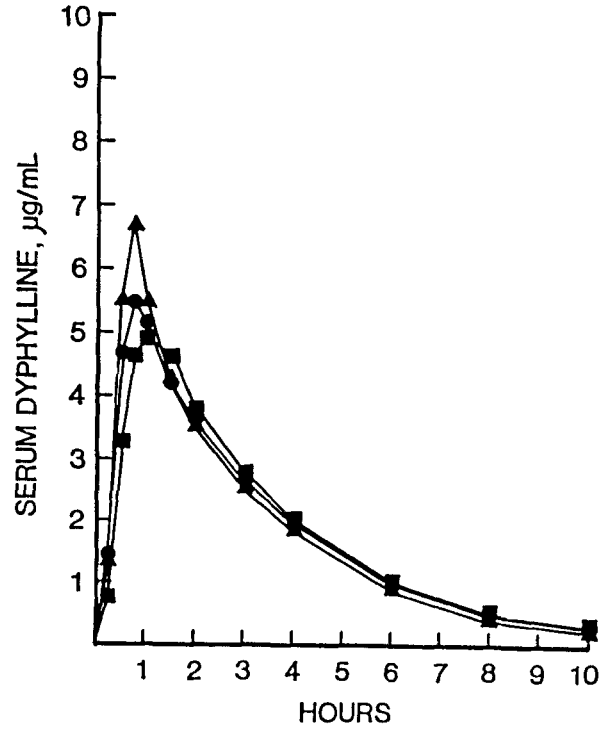

Figure 2—*Mean (n = 12) serum dyphylline concentrations after single doses (two tablets) containing 200 mg of dyphylline and 200 mg of guaifenesin. Key: (■) product 2; (▲) product 4; (●) product 6.*

and analyzed by direct injection, using the same HPLC system employed for the plasma assays.

Results and Discussion

Serum Concentrations at Each Time—Table I summarizes mean serum concentrations at each sampling time for the six dyphylline formulations. Figures 1 and 2 compare the mean serum concentrations for the three tablets containing dyphylline and dyphylline in combination with guaifenesin, respectively. The analysis of variance indicated significant differences ($p < 0.05$) among the six formulations only for the 0.25-, 0.5-, and 0.75-h samples. These data indicated a trend toward lower serum dyphylline concentrations during the early sampling times for the three tablets which also contained guaifenesin. However, except for the 8-h sample, where a 22% difference was observed, the differences in serum concentrations 1–10 h after dosing were <15% among the six tablets.

Bioavailability Parameters—Table II summarizes mean values for the various bioavailability parameters, and the significant differences noted with the Newman–Keuls analysis are given in Table III. There were no significant differences ($p > 0.05$) observed for the AUC values, the rate constants, or the half lives among the six products. The mean half life, which was ~2 h with all dosage forms, is consistent with previously reported values in healthy subjects.[2,3] The results of the power analysis indicated that 12 subjects were adequate to detect a difference of 28% for C_{max} and 17% for $AUC_{0-\infty}$) as statistically significant. The data in Table II indicate a trend toward lower bioavailability for the three products containing guaifenesin, although most differences were not statistically significant. This may be seen from the longer time to achieve maximum serum concentration, lower peak serum concentration, lower AUC, and longer mean residence time. However, the differences among the six tablets were only 7–12% for AUC and MRT, and none of the AUC differences were statistically significant. The reason for the tendency of the combination products to exhibit a slower and lesser extent of absorption is not known. Based on the study of Simons et al.,[5] who determined that aqueous solutions of dyphylline and dyphylline plus guaifenesin exhibited very similar absorption properties, it would seem that the results cannot be interpreted as resulting from a drug–drug interaction affecting absorption. The extent to which formu-

Table II—Mean Bioavailability Parameters[a]

Product	C_{max}, μg/mL	t_{max}, h	AUC_{0-10h}, μg·h/mL	k, h^{-1}	$t_{1/2}$, h	$AUC_{0-\infty}$, μg·h/mL	MRT, h
1	8.21	0.56	19.52	0.333	2.13	20.16	3.06
	(25)	(28)	(20)	(16)	(16)	(21)	(12)
2	5.80	1.09	18.26	0.336	2.08	18.38	3.48
	(22)	(32)	(14)	(10)	(10)	(20)	(10)
3	7.80	0.67	19.55	0.341	2.08	20.39	3.11
	(23)	(46)	(17)	(15)	(15)	(18)	(13)
4	6.96	0.75	18.29	0.358	1.98	19.06	3.12
	(24)	(38)	(15)	(19)	(14)	(17)	(13)
5	8.15	0.67	19.79	0.349	2.04	20.51	3.09
	(28)	(46)	(18)	(15)	(14)	(20)	(16)
6	6.17	0.98	18.60	0.345	2.04	19.31	3.30
	(22)	(71)	(18)	(11)	(12)	(18)	(16)
Percent Difference[b]	29.4	48.6	7.7	7.0	7.0	10.4	12.1

[a] Each value represents the mean of the 12 subjects. The RSD values are given in parentheses. [b] (Highest − Lowest)(100)/(Highest).

Table III—Newman-Keuls a Posteriori Test for Significant Product Differences at Each Sample Time

Observations	Product Ranking (Lowest to Highest)[a]					
Conc., 0.25 h	2	4	6	5	3	1
Conc., 0.50 h	2	6	4	3	5	1
Conc., 0.75 h	2	6	3	5	4	1
C_{max}	2	6	4	3	5	1
t_{max}	1	5	3	4	6	2
MRT	1	5	3	4	6	2

[a] Products underlined by a common line were not found to differ significantly ($p > 0.05$).

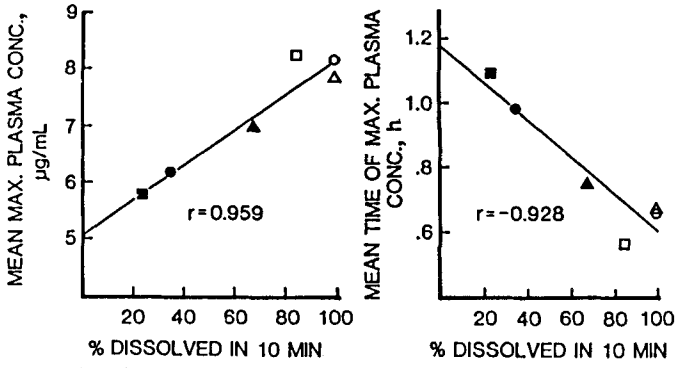

Figure 3—*Mean percent dissolved (n = 3) in vitro versus mean (n = 12) in vivo values for C_{max} and t_{max}. Key: (□) product 1; (■) product 2; (△) product 3; (▲) product 4; (○) product 5; (●) product 6.*

lation differences between the dyphylline and the dyphylline combination products can explain the observed differences cannot be determined from the available information.

Based on the <20% difference in AUC and C_{max}, and t_{max} differences of 0.5 h or less, it can be concluded that the three dyphylline formulations are bioequivalent, as are the three dyphylline–guaifenesin formulations.

In Vitro–In Vivo Correlation—Attempts were made to correlate the percent of drug dissolved in vitro at the various sampling times to the bioavailability parameters of $AUC_{0-\infty}$, C_{max}, and t_{max}. The parameters resulting in the highest correlation coefficients were observed for the mean percent of drug dissolved at 10 min and either the mean values for C_{max} or t_{max}, as illustrated in Fig 3. The RSD values ranged from 0.3 to 14.9% for the mean percent dissolution. The three tablet formulations which did not contain guaifenesin exhibited the shortest time to achieve the maximum plasma dyphylline concentration, the highest maximum dyphylline concentrations, and the most rapid dissolution.

References and Notes

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Acknowledgments

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