CHAPTER IV

RESULTS AND DISCUSSION

In Vitro Studies

All four commercial brands of gemfibrozil capsules were analyzed following the requirements as specified in the official monograph of the United States Pharmacopoeia XXII (United States Pharmacopoeial Convention Inc., 1990). Results of all tests are shown in Table 2. Each of them met the requirements for weight variation within the range of limit weight (± 15%). The content of active ingredient of all assayed products were within the limits of 90.0-110.0 percent labeled amount.

Disintegration time is an important attribute of capsule quality control because disintegration must take place before the active ingredient of the capsule can dissolve and be absorbed. All brands disintegrated within 15 minutes as specified in the United States Pharmacopoeia XXII (United States Pharmacopoeial Convention Inc.,1990) as general requirement for hard capsule. Disintegration time ranged from 7.86 to 9.44 minutes. The rank order of mean disintegration times were brands B > A > C > D. Statistical comparison, as shown in Tables 3 and 4, indicated that only the disintegration time of brand B was longer than that of brand A ($p < 0.05$) meanwhile

those of the other two brands (C and D) was not. However, the differences in disintegration times of drug-products appeared to be less importants for drug availability since it was just a process of drug disolution.

Dissolution testing is a crucial factor for systemic drug availability because a drug must, in general, dissolve to solution before being absorbed. Figure 2 and Table 5 illustrate the dissolution profiles of all four commercial brands of gemfibrozil capsules in phosphate buffer (pH 7.5 $\pm$ 0.1). Each brand reached the equilibrium state within about 90 minutes. The mean percent dissolved of gemfibrozil from all brands range from 100.55 to 102.01 at 180 minutes. The dissolution rate constants (K_d) were calculated from the slope of the first order plot between the amount of undissolve gemfibrozil versus time in semi-logarithmic scale. The dissolution rate constants of all brands are reported in Table 6. The rank order of all brands in term of dissolution rate constant were brands C > D > B > A. Statistical comparison, as shown in Tables 7 and 8, indicated that only the dissolution rate constant of brand B did not show statistically significant difference ($p > 0.05$) when compared to that of brand A where as the other two brands (C and D) did.

No statistical correlation ($p > 0.05$) was found between the disintegration times and the dissolution rate constants of all brands as shown in Table 9.

Table 2 In vitro Studies of Four Commercial Brands of Gemfibrozil Capsules

Brand	Weight[a] (mg)	% Labeled amount[b]	Disintegration Time[c] (min)
A	479.92 ± 0.02	98.25 ± 1.69	8.13 ± 0.16
B	486.61 ± 0.02	99.67 ± 3.78	9.44 ± 0.34
C	397.03 ± 0.01	105.33 ± 2.17	7.99 ± 0.22
D	388.89 ± 0.02	101.95 ± 2.73	7.86 ± 0.23

a = mean ± standard deviation (n = 10)

b = mean ± standard deviation (n = 3)

c = mean ± standard deviation (n = 6)

Table 3 Analysis of Variance for Disintegration Time of Four Commercial Brands of Gemfibrozil Capsules

Source of Variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	9.58	3.19	53.22
Within group	20	1.23	0.06	
Total	23	10.81		

$F^{e}_{0.05\ (3,20)} = 3.10$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 4 Comparison of Disintegration Time of Locally Manufactured Brands with that of Innovator's Product (Brand A) Using t-test

Brand	t (Calculated)	Statistical significance
B	9.26	S
C	1.00	NS
D	1.86	NS

$t^{a}_{(0.05,20)} = 2.09$

S = Significant ($p < 0.05$)

NS = Not significant ($p > 0.05$)

a = t-value obtained from the table

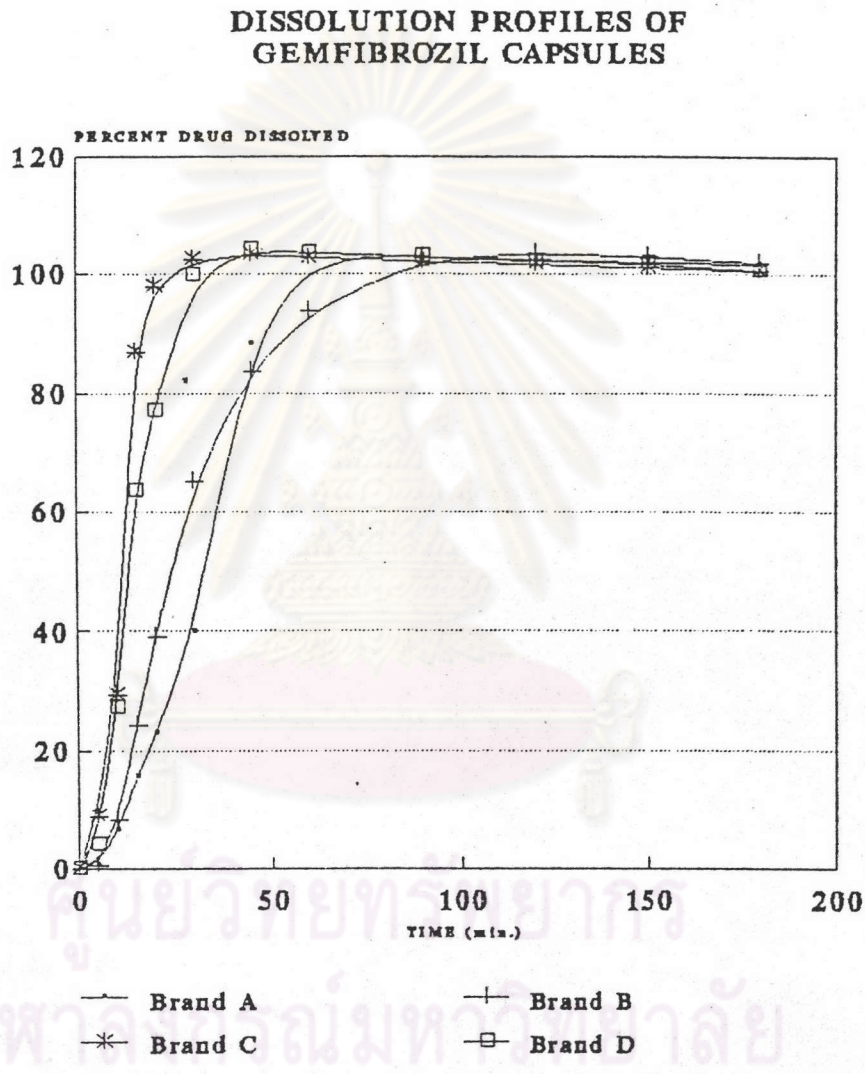

Figure 2 Dissolution profile of four commercial brands of gemfibrozil capsules in phosphate buffer (pH 7.5 $\pm$ 0.1

Table 5 Dissolution Profiles of Four Commercial Brands of Gemfibrozil Capsules in Phosphate Buffer (pH 7.5 ± 0.1)

Time (min)	Average Percent Gemfibrozil Dissolved[a]			
	Brand A	Brand B	Brand C	Brand D
5	0.46±0.39	0.55±0.43	8.80±2.82	4.03±2.91
10	6.57±6.74	8.26±5.05	29.32±4.06	27.15±16.58
15	15.73±9.66	24.31±6.57	86.88±8.12	63.42±35.29
20	22.89±9.77	39.07±6.23	97.82±5.49	76.88±42.51
30	39.73±13.54	65.03±8.73	102.39±4.80	99.88±7.74
45	88.37±10.13	83.69±6.93	103.41±3.70	104.06±4.14
60	103.06±2.71	93.80±7.33	102.88±2.92	103.68±3.96
90	103.02±1.99	102.69±4.39	102.08±2.00	102.93±2.11
120	102.45±2.01	103.66±2.32	101.77±2.03	102.22±1.78
150	102.01±0.79	102.94±2.02	101.06±1.64	101.85±1.56
180	101.66±0.77	102.01±1.96	100.55±1.04	100.72±1.65

a = mean ± standard deviation (n = 6)

Table 6 Dissolution Rate Constants of Four Commercial Brands of Gemfibrozil Capsules in Phosphate Buffer (pH 7.5 ± 0.1)

Brand	Dissolution Rate Constant[a] (K_d) (hr^{-1})
A	1.89 ± 0.38
B	2.58 ± 0.68
C	8.78 ± 3.50
D	6.28 ± 2.42

a = mean ± standard deviation (n = 6)

Table 7 Analysis of Variance for Dissolution Rate Constants of Four Commercial Brands of Gemfibrozil Capsules in Phosphate Buffer (pH 7.5 $\pm$ 0.1)

Source of Variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	188.17	62.72	13.41
Within group	20	93.58	4.68	
Total	23	281.75		

$F^{e}_{0.05\ (3,20)} = 3.10$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 8 Comparison of Dissolution Rate Constants of Locally Manufactured Brands with that of Innovator's Product (Brand A) Using t-test

Brand	t (Calculated)	Statistical significance
B	0.55	NS
C	5.51	S
D	3.51	S

$t^{a}_{(0.05,20)} = 2.09$

S = Significant ($p < 0.05$)

NS = Not significant ($p > 0.05$)

a = t-value obtained from the table

Table 9 In Vitro Parameters Correlation

Correlation	Correlation Coefficient	t value	Statistical Significance
Disintegration Times versus Dissolution Rate Constants	-0.55	0.93	NS

$t^{a}_{0.05\ (2)} = 4.30$

NS = Not significant ($p > 0.05$)

a = t value obtained from the table

The major factors responsible for differences in disintegration times and dissolution characteristics of these drug-products were diferences of raw materials used and/or production processes.

In Vivo Studies

1. Plasma Gemfibrozil Concentration Analysis

Chromatograms of gemfibrozil and ibuprofen (internal standard) are shown in Figure 3. The retention times of gemfibrozil and ibuprofen (internal standard) are 5.10 and 3.21 minutes, respectively. No any interferent peaks due to the presence of plasma proteins and/or endogenous substances were observed. The method of analysis was validated by determining the within-run and between-run precisions. The percent coefficient of variations (% CV) in the within-run and between-run precisions were 0.65-5.57 and 1.11 - 3.89 as shown in Tables 10 and 11, respectively. The standard calibration curve of gemfibrozil plasma concentrations versus peak height ratio of gemfibrozil to internal standard was linear covered up to 60 mcg/ml as fitted by linear regression (Appendix B). The efficiency of the separation technique used was evaluated by calculating the percentage of recoveries and comparing the peak height obtained from spiked plasma to the peak height from standard solution which were directly injected into the HPLC. Results as shown in Table 12

indicated that the analytical method used was independent of concentration. The percentage recoveries of gemfibrozil and internal standard were in the range of 67.14-80.95 and 80.90-87.95 respectively.

2. Plasma Gemfibrozil Levels

The plasma level of gemfibrozil at each sampling time (ranged from 0.5 to 10 hours) after oral administration of two 300 mg gemfibrozil capsules of brands A, B, C and D are shown in Tables 13 to 16, respectively. The plasma gemfibrozil concentrations at the time before drug administration each time in all subjects were equal zero. The concentrations of the drug in some subjects at the time 0.5 hour after dosing were quite low. This was because the recovery of gemfibrozil was about 74%. Individual plasma gemfibrozil concentration-time profile of twelve subjects are shown graphically in Figures 4 to 15. Comparison of the mean plasma concentration-time profile of four brands are summarized in Figure 16.

3. Evaluation of Bioavailability

The pharmacokinetic parameters namely the peak plasma drug concentration, C_{max}, the time to peak plasma drug concentration, t_{max} and the area under the plasma drug concentration versus time curve, AUC, are employed to characterized the bioavailability of orally pharmaceutical

formulations after administration. These parameters respect the rate (C_{max},t_{max}) and the extent (AUC) of drug absorption into systemic circulation. They can be obtained by deriving from the plasma drug concentration - time curves. In the bioequivalent study, drug products that are pharmaceutical equivalence will be bioequivalent if they are not significant difference with respect to the rate and the extent of drug absorption (Shargel and Yu, 1980).

Looking at the plasma gemfibrozil concentration versus time profiles of all subjects, the plots appeared to be unusual. Thus, various means, including computer program had been used to analyzed the data to obtain the relevant pharmacokinetic parameters for bioavailability comparisons. Initial results revealed that data analysis using the CSTRIP computer program appeared to be over estimates especially the C_{max} and AUC values. This indicated that the CSTRIP could not be used with these characteristic data. Contrastly when conventional method (normal curve fitting) was used, results were more acceptable. Hence the principal pharmacokinetic parameters obtain by the latter principle will be used for the purposes in this study.

The peak plasma drug concentration, the time to peak plasma drug concentration and the area under the plasma drug concentration versus time curve of all brands observed from the the data (C_{max}, t_{max}) and calculated

using trapezoidal rule (AUC) were shown in Tables 17, 20 and 22, respectively.

3.1 Peak Plasma Drug Concentration (C_{max})

The observed mean peak plasma gemfibrozil concentrations from individual plasma data of each brand were 31.72 $\pm$ 1.53, 19.68 $\pm$ 1.14, 26.43 $\pm$ 2.69 and 30.92 $\pm$ 1.22 mcg/ml for brands A, B, C and D, respectively as shown in Table 17. The peak plasma gemfibrozil concentrations appeared to be independent of dissolutions of the drug but they seemed to be correlative well with absorption processes. This was evident by dissolution of gemfibrozil from brand A was significantly low but its C_{max} value was greater than others with higher dissolution rates. Another contribution for higher C_{max} value might be due to good formulation. The better formulation and/or production processes, the higher the drug could be absorbed into blood circulation. The rank order of this value was brands' A > D > C > B. Statistical comparison indicated that brands B and C had peak plasma drug concentrations significantly lower than that of the innovator's product ($p < 0.05$) as reported in Tables 18 and 19.

3.2 Time to Peak Plasma Drug Concentration (t_{max})

The observed time to peak plasma gemfibrozil concentration of each individual is presented in Table 20.

The average peak times were 1.83 ± 0.17, 2.33 ± 0.30, 2.13 ± 0.16 and 1.92 ± 0.18 hours for brands A, B, C and D, respectively. There were no statistical different among these values ($p > 0.05$) as shown in Table 21. It revealed that gemfibrozil from all brands was begun to be absorbed at the same time eventhough dissolution of the drug from brands A and B were significantly lower than the other two brands (C and D). Another reason was absorption processes were slower than dissolution processes.

3.3 Area Under the Plasma Drug Concentration Versus Time Curve (AUC)

The AUC calculated from trapezoidal rule of individual plasma data of each brand were 97.00 ± 4.79, 75.22 ± 6.57, 81.23 ± 4.13 and 94.96 ± 5.26 mcg.hr/ml. for brands A, B, C and D, respectively (Table 22). The rank order of this value was brands' A > D > C > B. Statistical comparison results in Tables 23 and 24 indicated that brands B and C had area under the plasma drug concentration versus time curve lower than that of the innovator's product ($p < 0.05$) meanwhile that of brand D was not.

In this study, the factors responsible for higher or lower AUC values were the same ones that affected the C_{max} values.

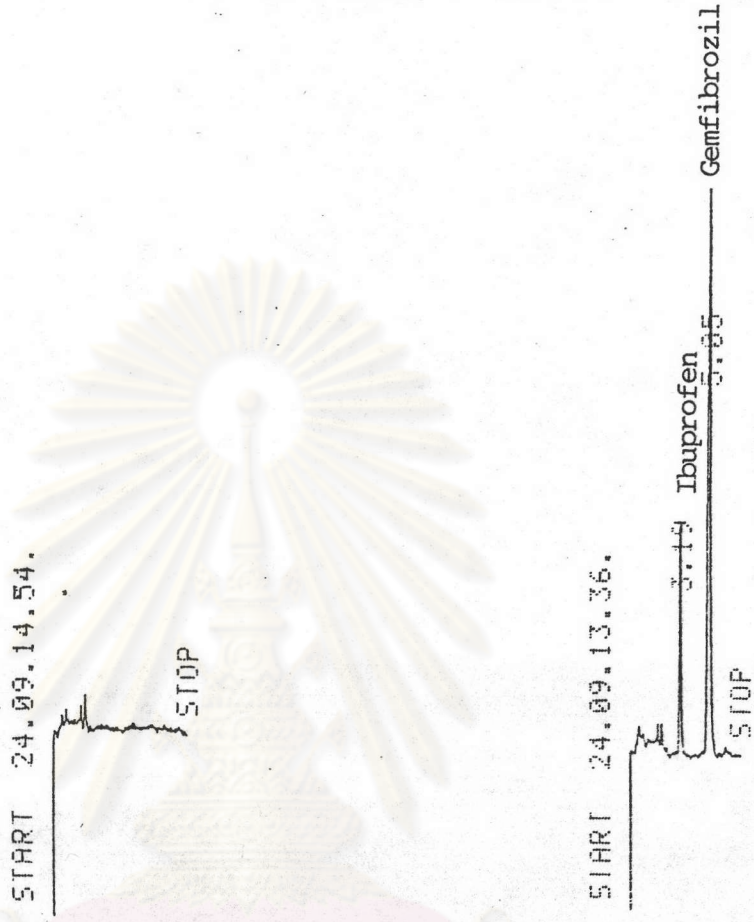

Figure 3 High pressure liquid chromatogram of plasma control (A) High pressure liquid chromatogram of gemfibrozil and ibuprofen (internal standard) (B

Table 10 Within-run Precision of Gemfibrozil from Three Replicated Plasma Calibration Curves Obtained in the Same Day

Concentration (mcg/ml)	Average* Peak Height Ratio	% CV
1.00	0.172	2.36
2.00	0.303	2.16
6.00	0.810	0.65
10.00	1.328	1.87
14.00	1.955	0.98
20.00	2.738	5.57
30.00	4.033	2.07
60.00	7.854	0.89

* n = 3

Table 11 Between-run Precision of Gemfibrozil from Three Replicated Plasma Calibration Curves Obtained in Three Different Days

Concentration (mcg/ml)	Average* Peak Height Ratio	% CV
1.00	0.172	1.43
2.00	0.304	2.93
6.00	0.809	1.33
10.00	1.355	3.89
14.00	1.911	2.05
20.00	2.653	2.74
30.00	3.960	2.02
60.00	7.840	1.11

* n = 3

Table 12 Recovery of Gemfibrozil and Internal Standard at Various Concentrations

Concentration (mcg/ml)	D[a] PH[c] (mm)		% Recovery	IS[b] PH[c] (mm)		% Recovery
	Solution	Plasma		Solution	Plasma	
1.00	9.50	6.50	68.48	45.5	38.5	84.62
2.00	13.0	10.0	76.92	45.0	36.5	81.11
10.00	35.0	23.5	67.14	43.0	35.0	81.40
14.00	52.5	42.5	80.95	41.5	36.5	87.95
20.00	100.5	76.5	76.12	44.5	36.0	80.90
60.00	127.0	100.0	78.74	42.5	37.0	87.06
Mean	-	-	74.73	-	-	83.84

a = Gemfibrozil

b = Ibuprofen (Internal standard)

c = Peak Height

d = % Recovery

$$= \frac{\text{Peak Height from Spiked Plasma}}{\text{Peak Height from Solution}} \times 100$$

Table 13 Plasma Gemfibrozil Concentrations (mcg/ml) from 12 Subjects Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Brand A

Subject No.	Time (hr.)								
	0.5	1.0	1.5	2.0	2.5	3.0	5.0	7.0	10.0
1	12.34	37.40	33.90	26.89	22.46	14.99	4.56	1.99	0.75
2	0.82	2.07	7.83	10.16	24.17	21.52	8.22	3.24	2.38
3	0.67	5.03	30.16	30.63	28.61	17.09	4.33	0.67	0.75
4	9.54	30.08	33.98	39.35	36.15	30.32	8.61	1.45	0.82
5	3.86	25.65	25.88	28.14	25.18	23.55	7.05	2.85	1.29
6	21.37	29.15	31.02	28.22	24.72	16.23	4.79	1.60	1.06
7	11.49	22.77	23.47	26.97	22.61	17.48	2.07	1.29	0.51
8	20.75	29.93	32.42	21.37	15.69	10.16	0.90	0.36	0.05
9	7.67	29.31	28.92	21.91	18.88	13.12	5.49	1.29	0.51
10	5.34	6.89	7.44	12.19	23.31	25.80	14.21	1.76	0.75
11	6.58	36.31	40.05	37.71	22.30	15.22	4.79	2.38	0.63
12	10.24	27.44	31.87	35.38	23.55	12.89	4.56	1.60	0.36
Mean	9.22	23.50	27.24	26.58	23.97	18.20	5.80	1.71	0.82
S.E.M.	1.93	3.49	2.90	9.10	1.44	1.71	1.00	0.24	0.17

Table 14 Plasma Gemfibrozil Concentrations (mcg/ml) from 12 Subjects Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Brand B

Subject No.	Time (hr.)								
	0.5	1.0	1.5	2.0	2.5	3.0	5.0	7.0	10.0
1	11.72	12.03	15.22	12.50	11.02	8.22	1.84	0.98	0.82
2	1.76	8.84	16.70	17.09	18.10	13.04	4.25	1.60	0.75
3	6.58	12.50	12.81	13.74	15.30	9.54	3.08	0.82	0.59
4	0.36	1.84	4.95	6.19	10.32	25.26	27.36	8.76	2.69
5	1.76	4.95	12.65	21.63	22.85	17.87	10.24	4.25	1.99
6	15.77	18.72	19.35	18.49	17.79	15.69	5.42	2.07	0.75
7	6.35	15.69	14.21	13.82	10.94	6.74	1.91	1.29	0.75
8	20.75	23.47	24.33	19.89	20.36	13.12	3.16	0.90	0.51
9	4.09	19.58	20.67	21.29	23.31	16.54	5.34	1.68	0.75
10	3.39	12.34	16.39	17.71	15.69	15.07	6.74	2.38	0.98
11	3.55	5.65	11.17	13.90	15.22	17.32	5.49	1.76	0.90
12	5.10	17.24	18.96	19.19	19.58	14.37	4.56	1.99	0.67
Mean	6.76	12.74	15.62	16.29	16.71	14.40	6.62	2.37	1.01
S.E.M.	1.79	1.89	1.46	1.28	1.28	1.43	2.00	0.69	0.19

Table 15 Plasma Gemfibrozil Concentrations (mcg/ml) from 12 Subjects Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Brand C

Subject No.	Time (hr.)								
	0.5	1.0	1.5	2.0	2.5	3.0	5.0	7.0	10.0
1	11.49	22.93	21.91	21.52	19.27	15.07	4.40	1.37	0.75
2	1.60	11.95	28.53	29.23	23.70	17.17	5.49	1.60	0.44
3	9.85	14.21	15.38	17.87	16.23	12.19	3.86	1.14	0.36
4	7.13	17.56	15.07	50.24	16.31	17.79	7.67	2.93	1.06
5	2.46	3.31	5.54	11.52	26.97	24.87	15.84	4.64	0.98
6	4.40	5.42	8.84	14.21	25.26	18.57	7.44	1.99	0.82
7	1.29	5.80	6.04	9.23	16.47	13.66	7.59	2.46	0.67
8	21.52	25.34	36.00	27.28	24.17	10.63	2.61	0.75	0.05
9	17.63	23.55	25.80	24.33	23.16	16.47	4.64	1.14	0.98
10	1.68	1.68	12.65	16.15	27.59	20.67	7.44	4.01	0.44
11	3.70	11.64	14.75	15.45	17.56	18.26	6.12	2.77	0.51
12	5.26	6.95	9.31	15.45	20.51	13.04	6.66	1.45	0.51
Mean	7.34	12.53	16.65	21.04	21.43	16.53	6.65	2.19	0.63
S.E.M.	1.91	2.39	2.75	3.19	1.22	1.44	0.96	0.35	0.09

Table 16 Plasma Gemfibrozil Concentrations (mcg/ml) from 12 Subjects Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Brand D

Subject No.	Time (hr.)								
	0.5	1.0	1.5	2.0	2.5	3.0	5.0	7.0	10.0
1	16.54	24.01	30.79	22.85	17.32	10.40	2.93	0.98	0.82
2	4.25	9.46	11.64	13.90	32.03	24.87	7.75	2.15	0.75
3	25.80	34.05	29.15	24.33	21.21	14.44	3.39	1.37	0.51
4	3.86	32.58	32.11	29.31	28.37	23.70	7.52	2.15	1.21
5	15.07	14.13	14.91	18.96	25.88	33.66	12.96	4.79	1.76
6	1.37	6.97	17.87	28.84	36.86	24.40	7.28	2.69	1.52
7	1.34	8.29	27.13	29.31	27.75	19.27	5.10	0.82	0.59
8	3.16	20.90	25.10	28.37	15.77	11.17	0.98	0.51	0.28
9	11.64	24.33	27.13	27.21	24.09	20.44	6.35	1.68	1.06
10	3.00	18.18	25.96	23.16	18.65	14.52	4.64	0.98	0.28
11	11.80	26.66	36.86	28.53	24.01	12.11	4.79	1.14	0.05
12	7.28	9.70	9.23	10.71	23.39	23.00	8.22	3.31	2.61
Mean	8.74	19.11	23.99	23.79	24.61	19.33	5.99	1.88	0.95
S.E.M.	2.19	2.74	2.49	1.82	1.77	2.03	0.90	0.36	0.21

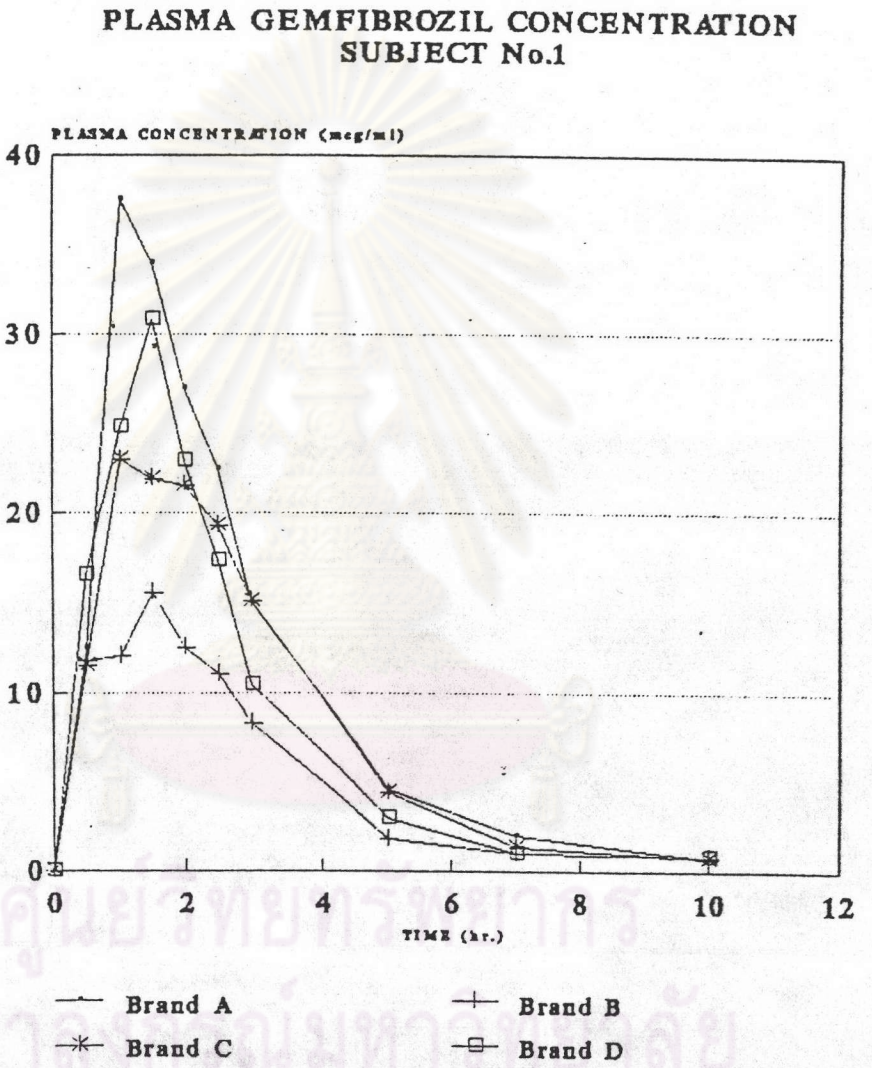

Figure 4 Plasma gemfibrozil concentration-time profile of subject No.1 following oral administration of two 300 mg gemfibrozil capsules

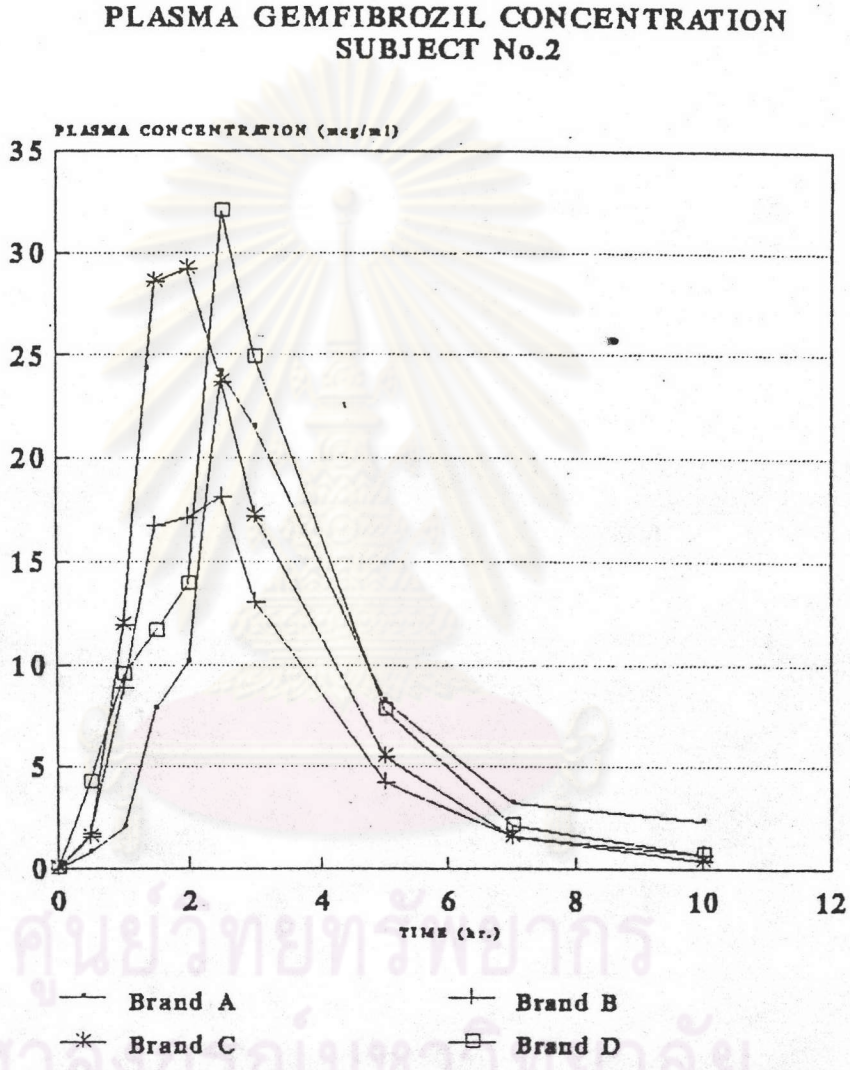

Figure 5 Plasma gemfibrozil concentration-time profile of subject No.2 following oral administration of two 300 mg gemfibrozil capsules

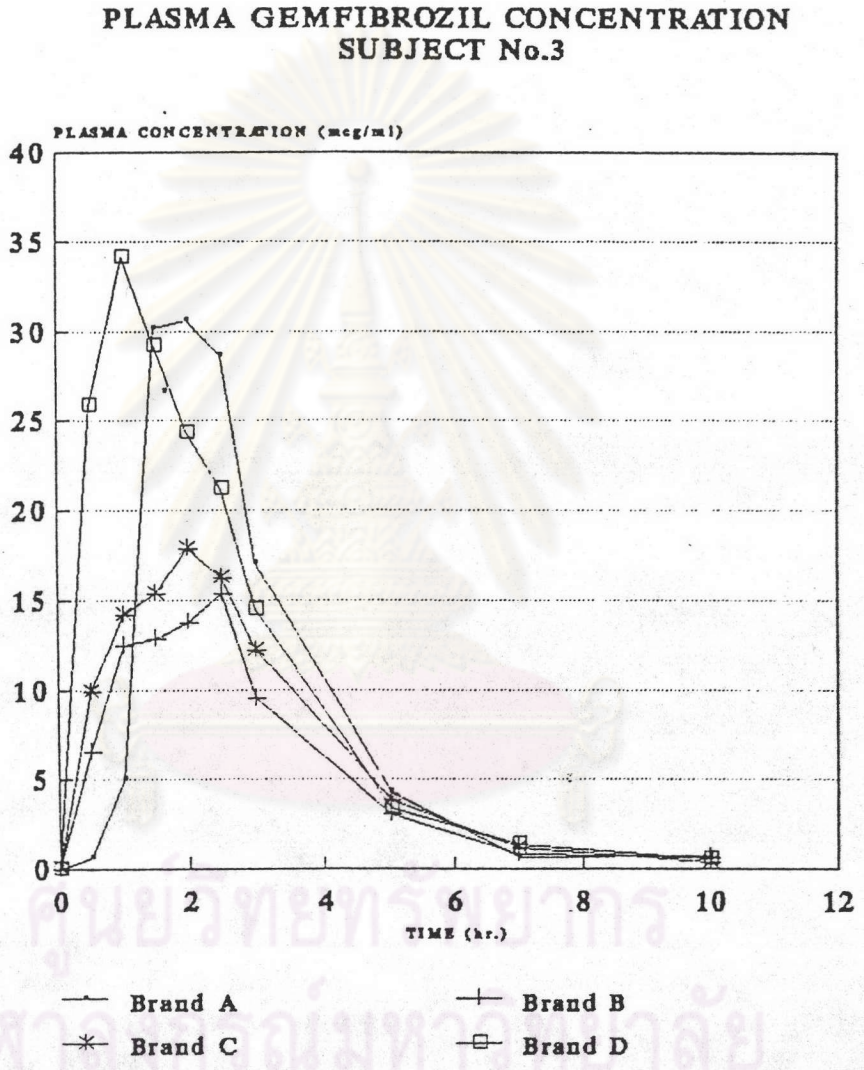

Figure 6 Plasma gemfibrozil concentration-time profile of subject No.3 following oral administration of two 300 mg gemfibrozil capsules

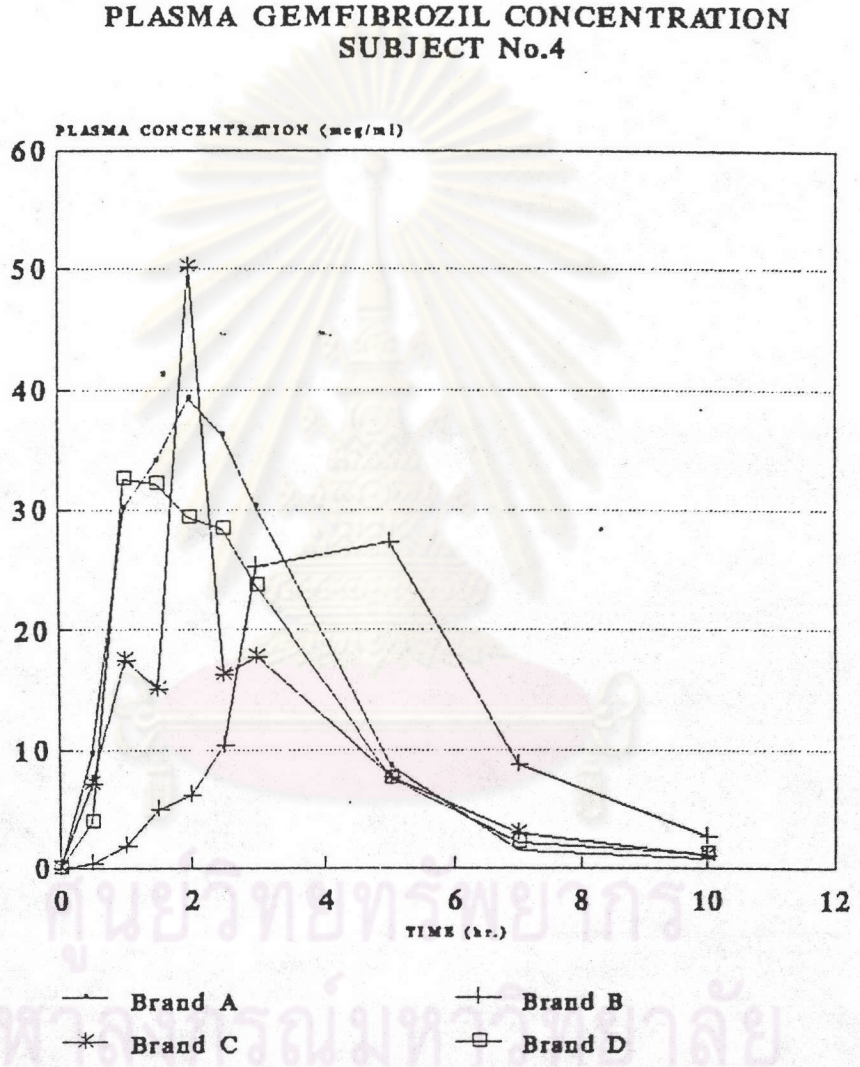

Figure 7 Plasma gemfibrozil concentration-time profile of subject No.4 following oral administration of two 300 mg gemfibrozil capsules

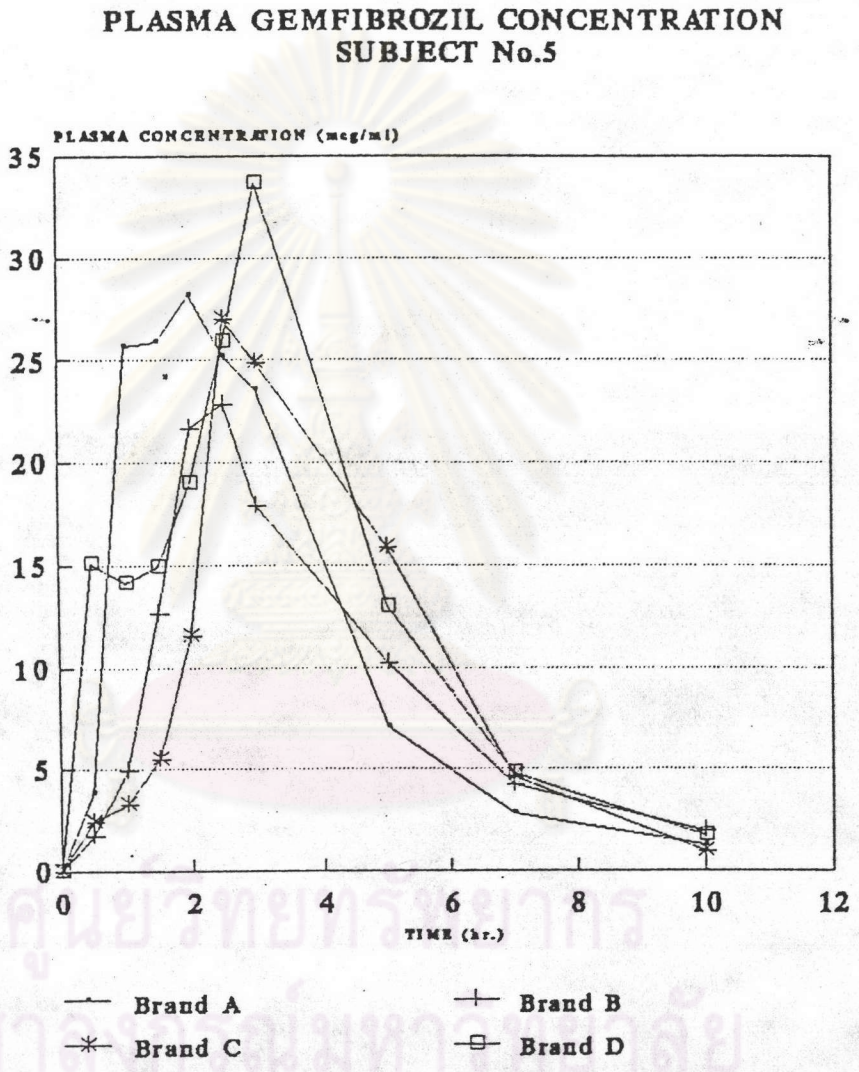

Figure 8 Plasma gemfibrozil concentration-time profile of subject No.5 following oral administration of two 300 mg gemfibrozil capsules

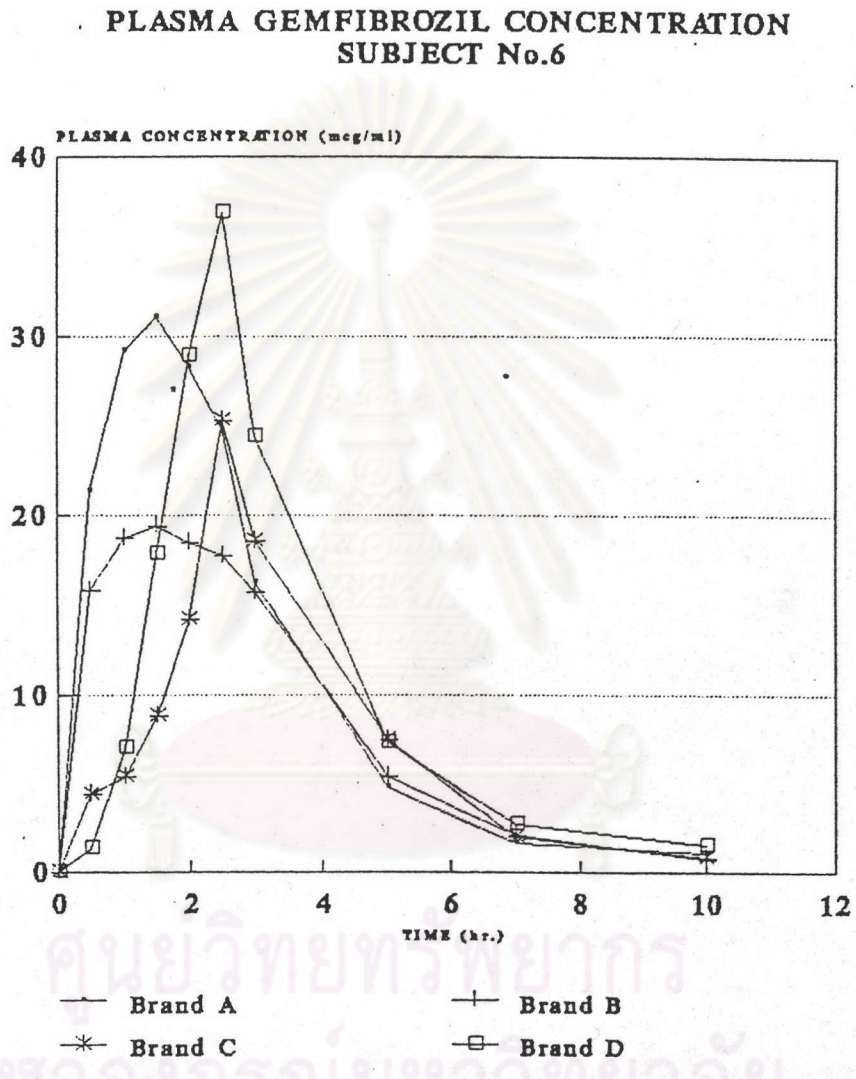

Figure 9 Plasma gemfibrozil concentration-time profile of subject No.6 following oral administration of two 300 mg gemfibrozil capsules

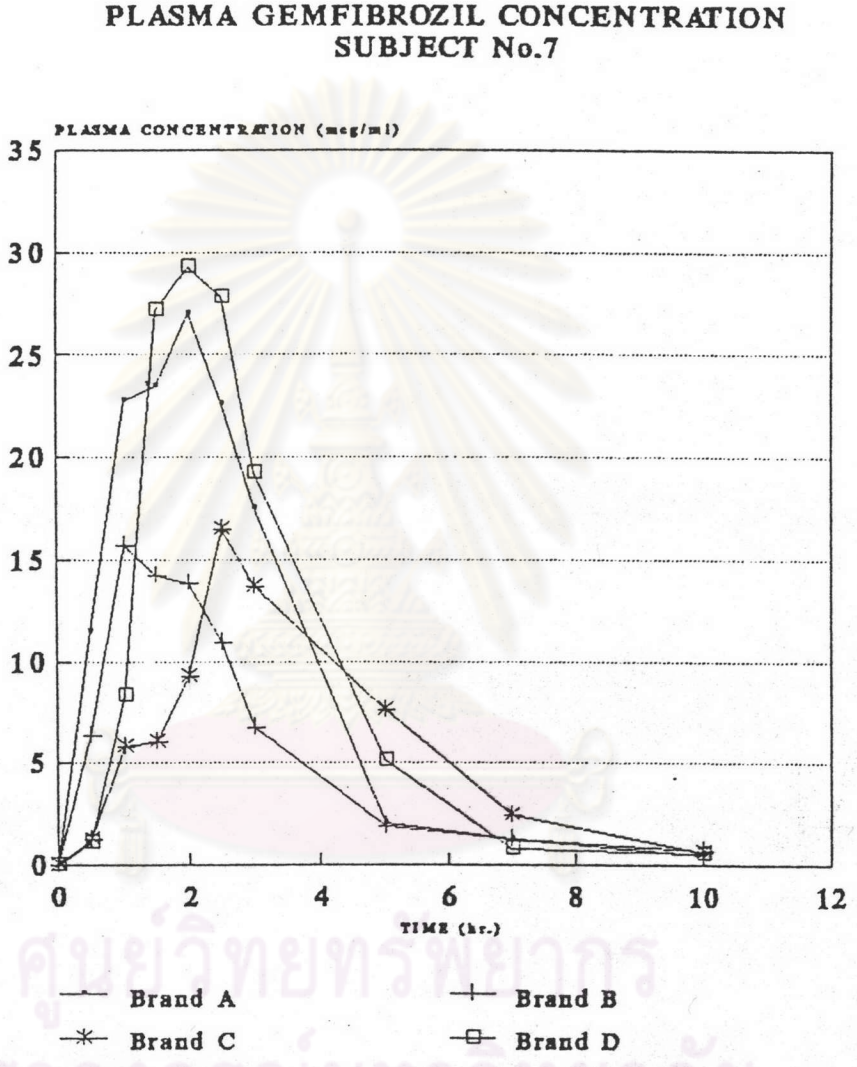

Figure 10 Plasma gemfibrozil concentration-time profile of subject No.7 following oral administration of two 300 mg gemfibrozil capsules

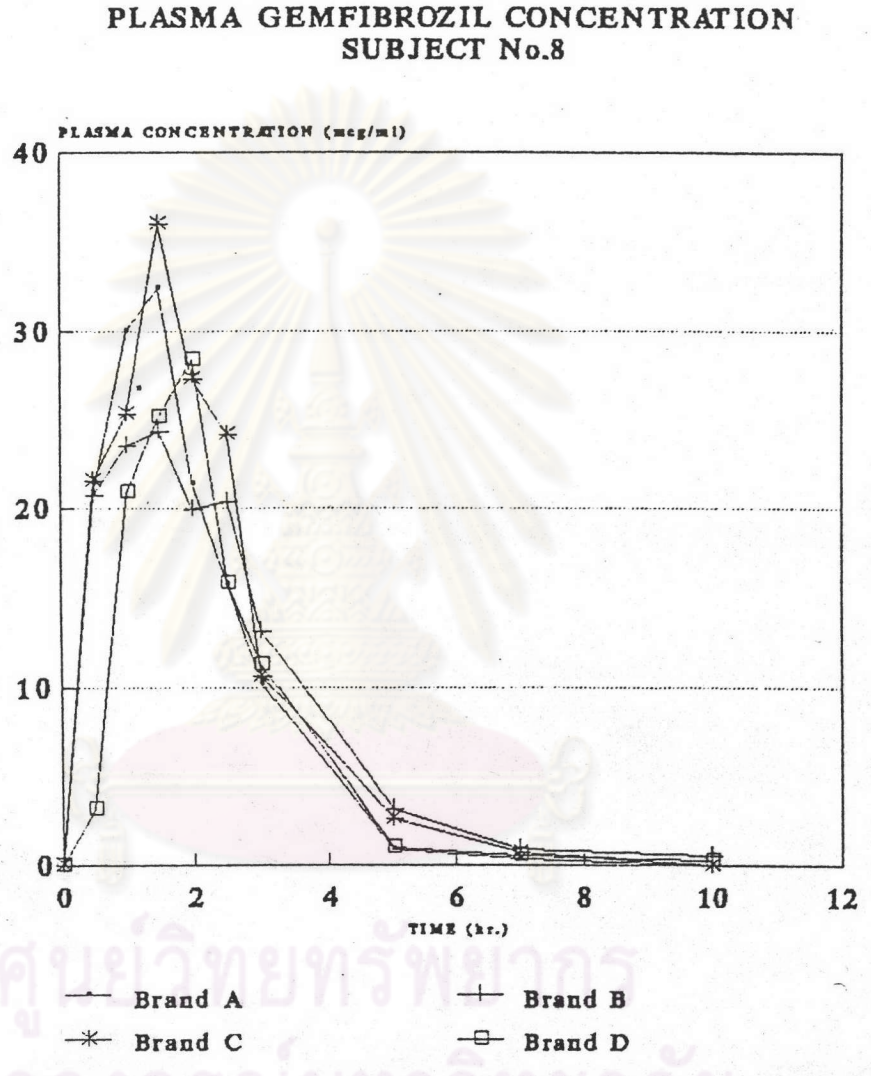

Figure 11 Plasma gemfibrozil concentration-time profile of subject No.8 following oral administration of two 300 mg gemfibrozil capsules

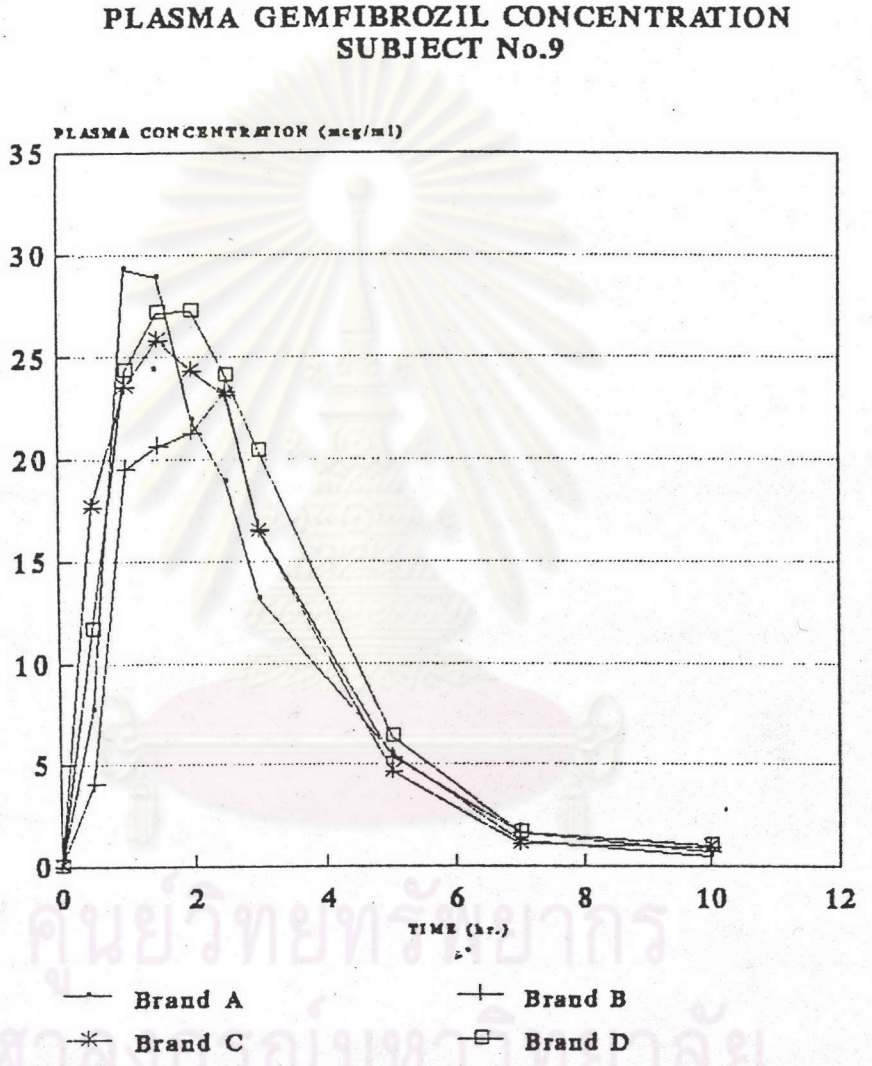

Figure 12 Plasma gemfibrozil concentration-time profile of subject No.9 following oral administration of two 300 mg gemfibrozil capsules

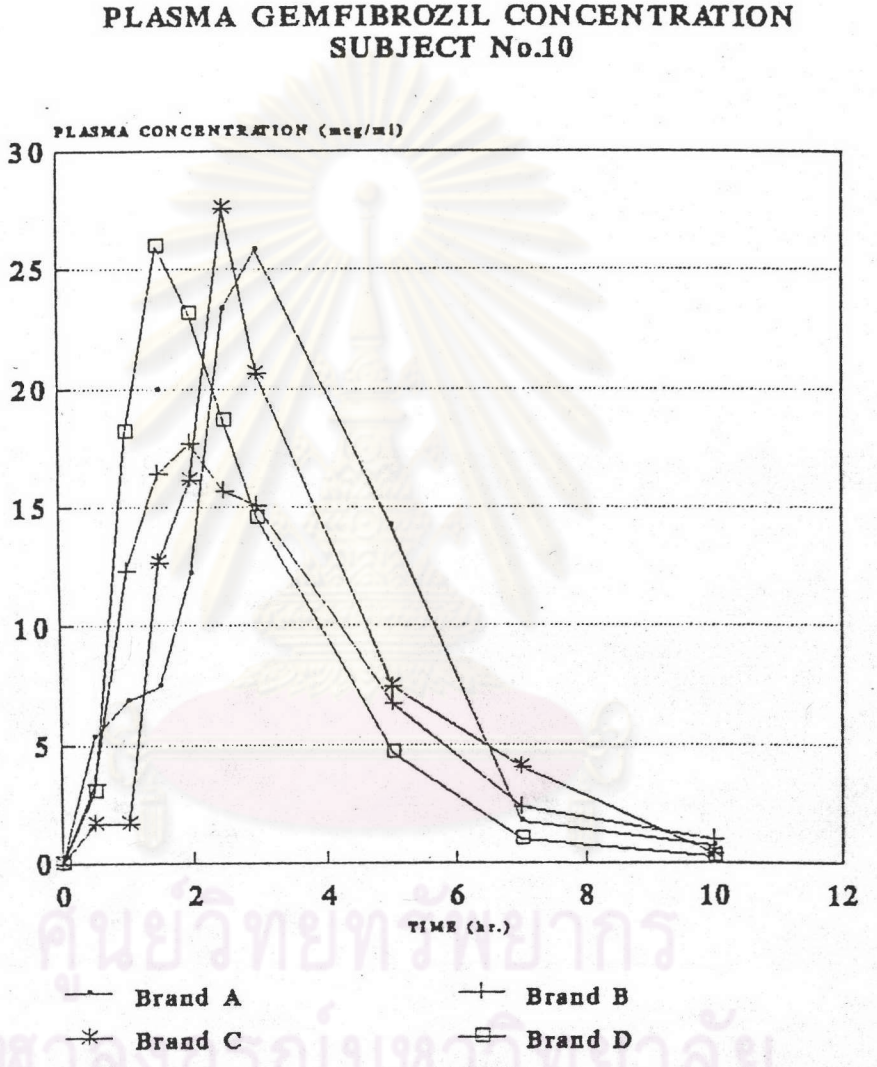

Figure 13 Plasma gemfibrozil concentration-time profile of subject No.10 following oral administration of two 300 mg gemfibrozil capsules

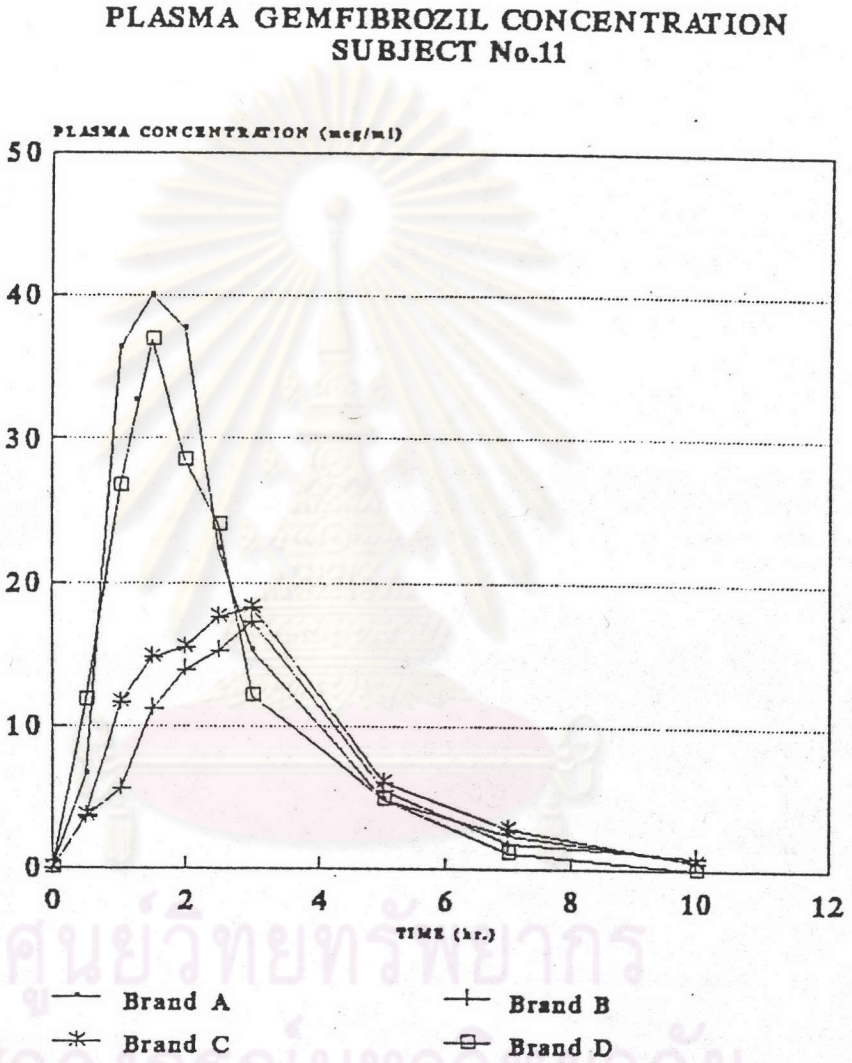

Figure 14 Plasma gemfibrozil concentration-time profile of subject No.11 following oral administration of two 300 mg gemfibrozil capsules

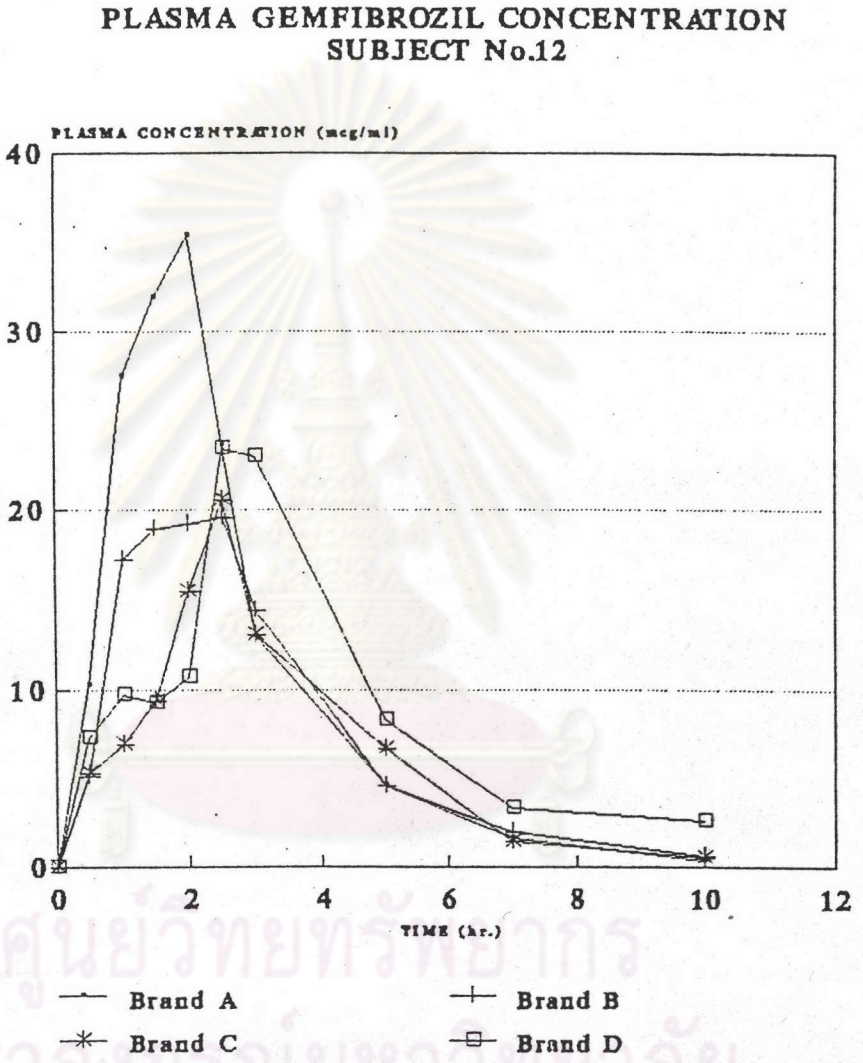

Figure 15 Plasma gemfibrozil concentration-time profile of subject No.12 following oral administration of two 300 mg gemfibrozil capsules

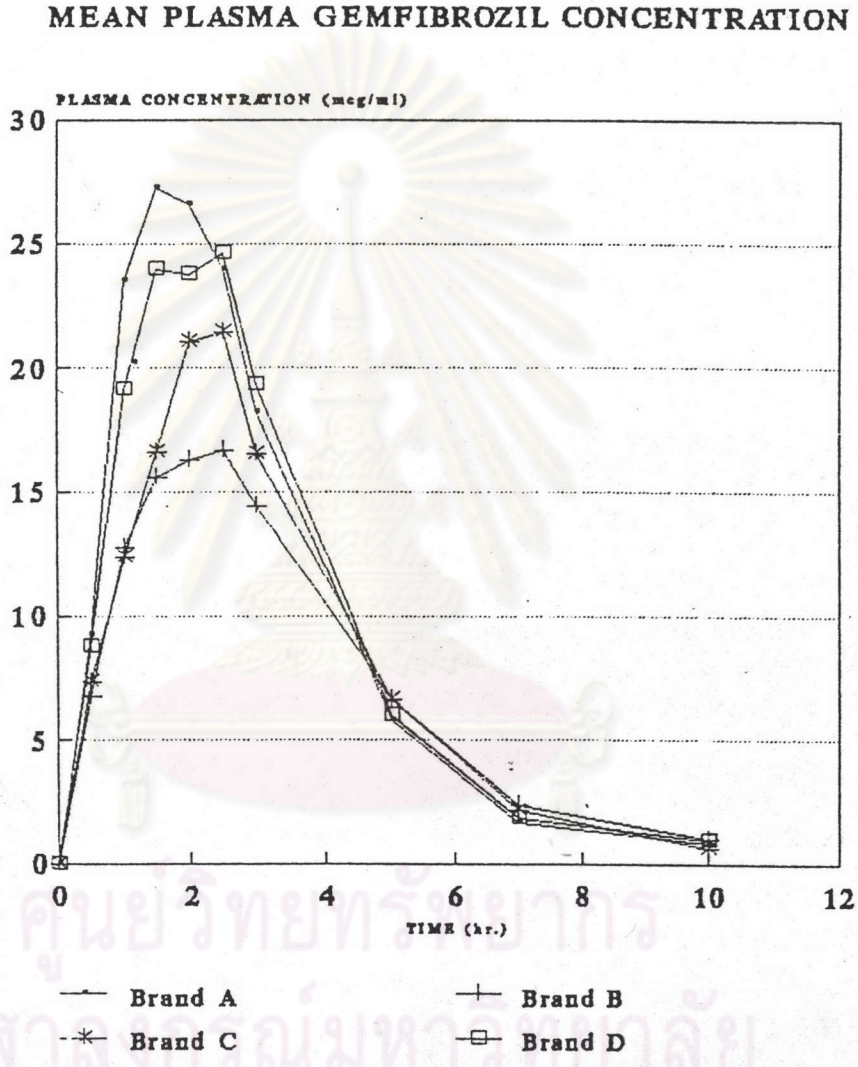

Figure 16 Comparison of mean plasma gemfibrozil concentration-time profile from 12 subjects following oral administration of two 300 mg gemfibrozil capsules of four commercial brands

Table 17 Peak Plasma Gemfibrozil Concentrations (C_{max}) Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	C_{max} (mcg/ml)			
	A	B	C	D
1	37.40	15.26	22.93	30.79
2	24.17	18.10	29.23	32.03
3	30.63	15.30	17.87	34.05
4	39.35	27.36	50.24	32.58
5	28.14	22.85	26.97	33.66
6	31.02	19.35	25.26	36.86
7	26.97	15.69	16.47	29.31
8	32.42	24.33	36.00	28.37
9	29.31	23.31	25.80	27.21
10	25.80	17.71	27.59	25.96
11	40.05	17.32	18.26	36.86
12	35.38	19.58	20.51	23.39
Mean	31.72	19.68	26.43	30.92
S.E.M.	1.53	1.14	2.69	1.22

Table 18 Analysis of Variance for Peak Plasma Gemfibrozil Concentrations of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	1,097.81	365.94	9.87
Within group	44	1,631.86	37.09	
Total	47	2,729.67		

$F^{e}_{0.05\ (3,44)} = 2.82$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 19 Comparison of Peak Plasma Gemfibrozil Concentrations of Locally Manufactured Brands with that of Innovator's Product (Brand A) Using t-test

Brand	t (Calculated)	Statistical Significance
B	4.84	S
C	2.12	S
D	0.32	NS

$$t^{a}_{0.05\ (44)} = 2.02$$

S = Significant ($p < 0.05$)

NS = Not significant ($p > 0.05$)

a = t-value obtained from the table

Table 20 Time to Peak Plasma Gemfibrozil Concentrations (t_{max}) Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	t_{max} (hr.)			
	A	B	C	D
1	1.0	1.5	1.0	1.5
2	2.5	2.5	2.0	2.5
3	2.0	2.5	2.0	1.0
4	2.0	5.0	2.0	1.0
5	2.0	2.5	2.5	3.0
6	1.5	1.5	2.5	2.5
7	2.0	1.0	2.5	2.0
8	1.5	1.5	1.5	2.0
9	1.0	2.5	1.5	2.0
10	3.0	2.0	2.5	1.5
11	1.5	3.0	3.0	1.5
12	2.0	2.5	2.5	2.5
Mean	1.83	2.33	2.13	1.92
S.E.M.	0.17	0.30	0.16	0.18

Table 21 Analysis of Variance for Time to Peak Plasma Gemfibrozil Concentrations of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	1.81	0.60	1.13
Within group	44	23.31	0.53	
Total	47	25.12		

$$F^{e}_{0.05\ (3,44)} = 2.82$$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 22 Area Under the Plasma Gemfibrozil Concentration Versus Time Curve (AUC) Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	AUC (mcg.hr./ml.)			
	A	B	C	D
1	102.05	50.98	82.49	80.08
2	84.97	62.91	85.42	90.36
3	81.79	52.06	63.81	97.29
4	136.05	130.96	102.37	117.66
5	110.02	94.38	102.78	131.28
6	105.06	83.51	75.15	103.55
7	84.64	49.07	60.22	85.21
8	75.63	81.16	87.67	64.73
9	85.76	82.75	93.60	103.60
10	95.17	74.95	82.11	75.26
11	108.24	65.27	75.33	91.64
12	94.65	74.59	63.77	98.89
Mean	97.00	75.22	81.23	94.96
S.E.M.	4.79	6.57	4.13	5.26

Table 23 Analysis of Variance for Area Under the Plasma Gemfibrozil Concentration Versus Time Curve of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	4,027.29	1,342.43	4.04
Within group	44	14,627.06	332.43	
Total	47	18,654.35		

$$F^{e}_{0.05\ (3,44)} = 2.82$$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 24 Comparison of Area under the Plasma Gemfibrozil Concentration Versus Time Curve of Locally Manufactured Brands with that of Innovator's Product (Brand A) Using t-test

Brand	t (Calculated)	Statistical Significance
B	2.93	S
C	2.12	S
D	0.27	NS

$$t^{a}_{0.05\ (44)} = 2.02$$

S = Significant ($p < 0.05$)

NS = Not significant ($p > 0.05$)

a = t-value obtained from the table

4. The Relative Bioavailability

Relative bioavailability is the availability of a drug product as compared to a recognized standard (innovator's product) (Shargel and Yu, 1980). In this study the relative bioavailability calculated using the mean AUC of each brand to that of the innovator's product. The values obtained for brands B, C and D relatively to that of brand A were 77.55, 83.74 and 97.90, respectively.

5. Evaluation of Bioequivalence

The principal pharmacokinetic parameters of gemfibrozil following oral administration of four commercial brands were summarized in Table 31. Statistical analysis of these corresponding parameters among the four commercial brands demonstrated that only brand D was completely bioequivalent to brand A in terms of both the rate and the extent of drug absorption whereas brands B and C were not.

6. Pharmacokinetic of Gemfibrozil Capsules

After analyzing the plasma gemfibrozil concentration-time relationship (Appendix E), results obtained demonstrated that the data were well described by means of biexponential equation. This referred that pharmacokinetic of gemfibrozil in Thai healthy volunteers could be explained by a one compartment open model.

The pharmacokinetic parameters derived from the model of analysis from plasma concentration-time data of each brand for gemfibrozil capsules were detailed in Tables 25 to 30.

6.1 Absorption Rate Constant (Ka)

The average absorption rate constants obtained from individual plasma data of brands A, B, C and D were 1.80 $\pm$ 0.19, 1.40 $\pm$ 0.12, 1.24 $\pm$ 0.13 and 1.53 $\pm$ 0.16 hr^{-1}, respectively (Table 25). There were no statistical difference among these values ($p > 0.05$) as shown in Table 26.

6.2 Elimination Rate Constant (K_{el})

The average elimination rate constants obtained from individual plasma data of brands A, B, C and D were 0.51 $\pm$ 0.03, 0.42 $\pm$ 0.01, 0.51 $\pm$ 0.03 and 0.50 $\pm$ 0.04 hr^{-1}, respectively (Table 27). These values were not statistical different from each other ($p > 0.05$) as seen in Table 28.

6.3 Biological Half-life ($t_{1/2}$)

The mean biological half-life of gemfibrozil determined for brands A, B, C and D were 1.41 $\pm$ 0.09, 1.67 $\pm$ 0.06, 1.40 $\pm$ 0.06 and 1.52 $\pm$ 0.15 hours, respectively (Table 29). The values agreed with those reported by Smith (1976) (1.5-2.0 hours). There were no statistical difference among these values ($p > 0.05$) as shown in Table 30.

Table 25 Absorption Rate Constants (K_a) of Gemfibrozil Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	K_a (hr^{-1})			
	A	B	C	D
1	3.36	1.86	1.71	2.02
2	0.61	1.31	1.43	0.82
3	1.75	2.10	1.68	2.76
4	1.65	0.63	1.15	1.79
5	1.87	1.17	0.81	0.81
6	1.82	1.34	0.80	1.24
7	1.83	1.19	0.94	1.34
8	1.88	1.84	1.34	1.66
9	2.15	1.44	2.22	1.55
10	0.89	1.38	0.81	1.68
11	1.93	0.99	1.03	1.67
12	1.83	1.57	0.96	0.97
Mean	1.80	1.40	1.24	1.53
S.E.M.	0.19	0.12	0.13	0.16

Table 26 Analysis of Variance for Absorption Rate Constants of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	1.98	0.66	2.36
Within group	44	12.23	0.28	
Total	47	14.20		

$F^{e}_{0.05\ (3,44)} = 2.82$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 27 Elimination Rate Constants (K_{el}) of Gemfibrozil Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	K_{el} (hr^{-1})			
	A	B	C	D
1	0.47	0.39	0.43	0.46
2	0.32	0.43	0.54	0.46
3	0.53	0.45	0.52	0.51
4	0.54	0.39	0.39	0.45
5	0.42	0.33	0.48	0.42
6	0.45	0.44	0.46	0.43
7	0.51	0.38	0.48	0.55
8	0.79	0.51	0.82	0.60
9	0.49	0.47	0.45	0.45
10	0.54	0.40	0.53	0.58
11	0.50	0.42	0.50	0.80
12	0.56	0.45	0.50	0.23
Mean	0.51	0.42	0.51	0.50
S.E.M.	0.03	0.01	0.03	0.04

Table 28 Analysis of Variance for Elimination Rate Constants of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	0.07	0.02	2.00
Within group	44	0.49	0.01	
Total	47	0.56		

$F^{e}_{0.05\ (3,44)} = 2.82$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 29 Biological Half-life ($t_{1/2}$) of Gemfibrozil Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Subject No.	$t_{1/2}$ (hr)			
	A	B	C	D
1	1.48	1.79	1.60	1.49
2	2.18	1.62	1.28	1.51
3	1.30	1.54	1.33	1.37
4	1.27	1.76	1.77	1.55
5	1.66	2.11	1.45	1.63
6	1.56	1.59	1.52	1.60
7	1.35	1.83	1.44	1.26
8	0.87	1.37	0.85	1.15
9	1.40	1.48	1.54	1.55
10	1.27	1.74	1.30	1.20
11	1.39	1.67	1.38	0.86
12	1.23	1.54	1.39	3.06
Mean	1.41	1.67	1.40	1.52
S.E.M.	0.09	0.06	0.06	0.15

Table 30 Analysis of Variance for Biological Half-life of Four Commercial Brands

Source of variation	d.f.[a]	SS[b]	MS[c]	F[d]
Among group	3	0.55	0.18	1.50
Within group	44	5.18	0.12	
Total	47	5.73		

$F^{e}_{0.05\ (3,44)} = 2.82$

a = Degree of freedom

b = Sum of square

c = Mean square

d = Variance ratio

e = F value obtained from the table

Table 31 Estimated Pharmacokinetic Parameters (Mean ± SEM) of Gemfibrozil From Twelve Healthy Volunteers Following Oral Administration of Two 300 mg Gemfibrozil Capsules of Four Commercial Brands

Parameters	Brand				Statistical Significance
	A	B	C	D	
C_{max} (mcg/ml)	31.72±1.53	19.68±1.14	26.43±2.69	30.92±1.22	S (A=D > B, C)
t_{max} (hr)	1.83±0.17	2.33±0.30	2.13±0.16	1.92±0.18	NS
AUC (mcg.hr/ml)	97.00±4.79	75.22±6.57	81.23±4.13	94.96±5.26	S (A=D > B, C)
K_a (hr^{-1})	1.80±0.19	1.40±0.12	1.24±0.13	1.53±0.16	NS
K_{el} (hr^{-1})	0.51±0.03	0.42±0.01	0.51±0.03	0.50±0.04	NS
$t_{1/2}$ (hr)	1.41±0.09	1.67±0.06	1.40±0.06	1.52±0.15	NS

S = Significant ($p < 0.05$)

NS = Not significant ($p > 0.05$)

7. In Vitro-In Vivo Correlation

The correlation studies between the in vitro and in vivo data for gemfibrozil brands A, B, C and D are presented in Table 32. No statistical correlation ($p > 0.05$) was found between the in vitro and in vivo parameters indicating that the in vitro parameters could not be used to predict the bioavailability of gemfibrozil.

Table 32 In Vitro-In Vivo Correlations

Correlation	Correlation Coefficient	t value	Statistical Significance
Disintegration Time versus C_{max}	- 0.89	2.76	NS
Disintegration Time versus t_{max}	0.80	1.88	NS
Disintegration Time versus AUC	- 0.73	1.51	NS
Dissolution Rate Constant versus C_{max}	0.11	0.16	NS
Dissolution Rate Constant versus t_{max}	0.06	0.08	NS
Dissolution Rate Constant versus AUC	- 0.14	0.20	NS

$t_{0.05\,(2)} = 4.30$

NS = Not significant ($p > 0.05$)

a = t value obtained from the table