LEBANESE AMERICAN UNIVERSITY
BYBLOS-LEBANON

A COMPARATIVE SINGLE-DOSE BIOEQUIVALENCE STUDY OF TWO GLIBENCLAMIDE BRANDS AMONG HEALTHY VOLUNTEERS

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ABSTRACT

Glibenclamide is a second generation sulfonylurea oral hypoglycemic agent that plays an important role in the therapy of type II diabetes mellitus (DM-II); moreover, glibenclamide (Glibamid®) is being extensively used among diabetics in the middle east, including Lebanon where this drug is manufactured, without any clinical in vivo implication showing or confirming its bioequivalency. So, this investigation was carried out to evaluate the in vitro dissolution as well as the bioavailability and pharmacokinetic properties of two tablet oral dosage forms of glibenclamide, Daonil® (drug A) and Glibamid® (drug B) in a single dose of 5 mg among healthy volunteers.

The two products were found to comply with the compendial requirments for both disintegration and content uniformity; moreover, the in vitro dissolution characteristics of the two products were similar.

Method: Ten healthy male volunteers were enrolled in the study, each received a single dose of each drug in an open randomizes two-way cross-over study, with a wash out period of 7 days. Blood samples were obtained over a 10 hours interval according to this fashion : At zero, 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 6, 8, and 10 hours. These samples were analyzed for serum glucose by the glucose oxidase method and glibenclamide by a sensitive HPLC assay.

Results: The two products were closely related in terms of their in vitro compendial requirements. Moreover, there was no significant difference with respect to peak serum concentration (103.92 ± 43.98 and 98.5 ± 51.26 ng/ml for products A and B, respectively) or to the corresponding peak times (2.6 ± 0.66 and 2.3 ± 0.88 hours for A and B respectively). Furthermore, the difference between area under serum concentration-time curve (AUC) for the two products (390.86 ± 152.61 and 360.7 ± 160.21 ng hr /ml for A and B, respectively) was not statistically significant, with $P > 0.05$. The comparable serum glucose levels for the two products supported the pharmacodynamical equivalence between the two glibenclamide brands.

Conclusion: The findings in this study indicates that the two products of glibenclamide are bioequivalent in terms of bioavailability and pharmacodynamic effect on healthy male volunteers.

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CHAPTER I

INTRODUCTION

Glibenclamide is a second generation sulfonylurea oral hypoglycemic agent that plays an important role in the oral therapy of type II diabetes mellitus (DM II). The precise mechanism(s) of hypoglycemic action of sulfonylurea antidiabetic agents has not been clearly established, however it initially appears to lower blood glucose concentration principally by stimulating secretion of endogenous insulin from the β-cells of the pancreas. Other mechanism of the hypoglycemic action associated with short-term glibenclamide therapy appears to include reduction of basal hepatic glucose production and enhancement of peripheral insulin action at post receptor sites.[1,2,3,4,5,6,7]

The usual initial dose of glibenclamide is 5 mg daily preferably with or immediately after the first main meal; 2.5 mg is used in debilitated or elderly patients.[1,2,3]

Peak glibenclamide plasma levels are usually reached within 2-6 hours after oral absorption, food usually does not alter the rate or completeness of glibenclamide absorption . Glibenclamide is 99 % bound to plasma protein; metabolized almost completely in the liver with the principle metabolite being only very weakly active. Approximately 50 % of a dose is excreted in the urine and the other 50 % via the bile into the feces. The half-life elimination is approximately 5-7 hours . As for adverse effects mainly hypoglycemia, gastrointestinal reactions: cholestatic jaundice and

hepatitis may occur rarely, liver function abnormalities including isolated transaminase elevation have been reported, dermatologic reactions: allergic skin reaction e.g pruritus, and erythema occurred in 1.5% of treated patients, photosensitivity reactions have also been reported, hematologic reactions: such as leukopenia, thrombocytopenia presented as pupura and agranulocytosis , hemolitic anemia and aplastic anemia have been also reported. Metabolic reactions: such as cases of hyponatremia have been reported with glibenclamide and all other sulfonylurea due to its diuretic effect. [2,3,8,9,10,12,13,14]

Glibenclamide is being extensively used among diabetics in the middle east, including Lebanon where this drug Glibamid® (glibenclamide) is being manufactured by Pharmaline (A Lebanese pharmaceutical industry), without any clinical in vivo implication showing or confirming its bioequivalency. Moreover, recent studies showed that commercialized glibenclamide products are not all equally bioavailable, due to the incomplete absorption by the human gut, thus accounting for large and unpredictable variation in blood levels following oral administration of the drug [8,9,12,14].

These reasons mentioned above, strongly support the need for a bioequivalency study between two different glibenclamide formulations (Glibamid®/Daonil®). Thus , the objective of this study is to compare the bioavailability and pharmacokinetic properties of two different glibenclamide formulations in a single oral dose of 5 mg, among healthy volunteers.

CHAPTER II

MATERIALS AND METHODS

Products

Drug "A" Daonil® tablets, 5 mg; lot # 41C818; Hoechst laboratories, Frankfurt, Germany.

Drug "B" Glibamid® tablets, 5 mg ; lot # 2851 1; Pharmaline laboratories, Nahir Ibrahim, Lebanon.

In vitro studies

Uniformity of content

The content uniformity of tablets were determined using the BP 98 method. Ten tablets of each brand were individually assayed for glibenclamide using a stability-indicating assay as outlined under the dissolution test.

Disintegration test

The disintegration time of both brands was determined using the BP 98 disintegration method for uncoated tablets. Ten tablets of each brand were used .

Dissolution test

The basket method of the USP XXI for uncoated tablets was used for the in vitro dissolution study. Five hundred milliliters of phosphate buffer pH 7.4 was agitated at 100 rpm and maintained at 37°c. Aliquots were withdrawn at predetermined time intervals (10, 30, 60, and 90 minutes) and filtered through a 0.45 µm membrane filter. Substitution with fresh buffer preheated to 37°c was done after each withdrawal. The filtered solution were assayed using the HPLC method where

20µl was injected at each time. The method is based on eluting properly diluted solutions of glibenclamide from an ultrasphere ODS 5 µm (4.6 x 250mm, C18) column with a filtered and degassed mobile phase consisting of a mixture of 47 volumes of acetonitrile and 53 volumes of a 1.36 % w/v solution of potassium dihydrogen orthophosphate, previously adjusted to pH 3 with orthophosphoric acid, at a flow rate of 2 ml/min. The eluent was monitored at 300nm wavelength (λ) and at a range of 0.01 aufs.

In vivo study

Subjects

Ten healthy male volunteers participated in an open, randomized two-way cross-over study. Their mean age was 25.2 ± 2.15 years with a range of (22 – 30 years), body weight of 82.6 ± 15.13 Kg with a range of (63 – 120 Kg) and height of 177.1 ± 9.8 cm with a range of (162 – 191 cm). One week prior to enrolment a complete physical exam and a biochemical screening was performed, (hematology, blood biochemistry including glucose and urine analysis)(table 1). Subjects with history or evidence of diabetes, cardiac, renal, hepatic, gastrointestinal disease, glucose intolerance or sulfonylurea allergy were excluded from the study. Volunteers were asked to abstain from taking any drug for at least one week prior to physical exam.

Informed consent, shown on page 23, was obtained from the volunteers after explaining the nature and purpose of the study, along with a brief explanation about glibenclamide effects.

Clinical study

Volunteers appeared at the Notre Dame De Secours Hospital (NDDSH), Byblos, Lebanon; on two consecutive Sunday mornings at 6:30 AM after being fasting for 10 hours before the study took place. Medications were given at 7:30 AM and blood samples were taken for a period of 10 hours thereafter. The study was divided into two periods where the two drugs were orally taken in a randomized cross-over order one each Sunday with a washout period of 7 days. At 7:15 AM, each subject received a 200 ml of orange juice with 7.5 gm of sugar; at 7:30 AM, the first blood sample was taken and each volunteer received a 5 mg single oral dose of either brands (A or B) with 200 ml of water. Breakfast was served 1.5 hours after initiation of therapy, a 200 ml of orange juice or coffee break at 4 hours, a standardized lunch at 6 hours, and a light snack at 8 hours. All volunteers were asked to abstain from strenous activities and smoking on the study day.

Blood samples (10 ml /drawing), were obtained from the cubital and forearm veins through an indwelling heparinized catheter, put into coded plain tubes at 0, 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 6, 8, and 10 hours post dosing. Each subject was called for blood withdrawal at the desired time interval monitored by our team members. The withdrawn blood (10 ml) was put in a labeled tubes each according to the volunteers

ID #, Time, and drug code; where thereafter sent to the lab, centrifuged at 5000 Hz for 10 minutes, after coagulation. Serum was separated and transferred into a 10 ml labeled tube where it was divided into two portions; one used for immediate glucose checkup and the second portion was stored at –20 °c pending drug analysis.

Analytical techniques

Serum glucose concentrations were determined using the glucose oxidase method.[10] While glibenclamide serum concentrations were determined using the highly specific reversed-phase, high performance liquid chromatography (HPLC) method with ultra violet (UV) detection.[11]

Instruments

A high performance liquid chromatography (HPLC) consisted of a dual pumps system (Waters, model 510), and an injector (Rheodyne) with a 200 µl loop size were used. The system was equipped with a variable ultraviolet detector (Waters, model 486), a chart recorder integrator (model 746, Waters Data module) and a spherisorb C_8 3.9x 150 mm. ID, particle size 5µm) phase separation, Nova-pak, Waters Inc , Ireland). The ultraviolet detector was set at a wavelength (λ = 230 nm) and sensitivity range of 0.01 aufs. The chart recorder (integrator) was adjusted at 2 mm/ min. And the system was put on 2 ml/min flow rate.

Materials and reagents

Pharmaceutical grade glibenclamide (Glibenclamide USP reference, lot # F-2, cat # 29550, Rockville), Flufenamic acid 97 % (lot # 47212/1, New jersey USA), and acetonitrile 190 for UV/ gradient quality (Romil Ltd, England), were used.

The mobile phase was prepared by mixing acetonitrile with deionized water (millipore) in a ratio of 45:55, with pH adjusting between 3.7 – 3.8 using glacial acetic acid and 2M sodium hydroxide solution. The mobile phase was degassed under vacuum using a

0.45 µm x 47 membrane filter (Nylon 66, Supalco Inc, Bellifonte) and a degassing system (Branson 2210) where, thereafter it was pumped through the column at a flow rate of 2 ml/min (1000 – 3000 psi; 10800 – 15000 Kpa) at ambient temperature.

Preparation of standards

An accurate weight of 10 mg glibenclamide was dissolved in 10 ml acetonitrile. A 100µl volume was transferred to a 10 ml volumetric flask and diluted to volume with the mobile phase to give a working standard solution (WSS) of 10 µg/ml. On the other hand, the working internal standard solution (WISS) was prepared by diluting 100µl of the stock solution (10mg flufenamic acid in 10ml acetonitrile, giving a solution of 1µg/µl) to 10ml with mobile phase to give a WISS of 10 µg/ml. Stock solutions prepared with acetonitrile were stable at 4°c for more than one month.

Analytical procedure

Accurate volumes of 50 µl glibenclamide WSS (10µg/ml) was added to 50µl volume of WISS in 1.5 ml microcentrifuge polypropylene (eppendrof) tube, where the mixture was vortexed for 30 sec, 50 µl of the mixture was injected into the HPLC column where the integrator revealed peaks for glibenclamide and flufenamic acid with retention times 4.0 and 6.5 minutes respectively (figure 1). As for the serum trial sample, we took an accurate volume of 20 µl of glibenclamide WSS (10µg/ml) we added 0.5 ml serum, and 25 µl of the WISS in a 10 ml screw-capped tubes. The mixture was vortexed for 30 sec. Two milliliters of acetonitrile were added as protein precipitant. The sample was vortexed again for 1 min and then centrifuged at 2500 Hz for 10 min. The clear supernatant was transferred to a centrifuge tube and evaporated to dryness in a water bath thermostatically controlled at 45°c under a gentle stream of dry nitrogen. The residue was taken up in 200 µl of mobile phase and transferred into a 1.5 ml microcentrifuge polypropylene (eppendrof) tube, and centrifuged for 2 min at 12,000. Hz in a microcentrifuge. An appropriate volume (50 – 70 µl) was injected into the HPLC system, and the result is reflected in figure 2.

As for the volunteers serum samples shown in figure 3, they were similarly treated, by adding 1.5 ml serum to 200 µl WISS (10µg/ml) in a 10 ml screw-capped tubes and the mixture was vortexed for 60 seconds. 4.5 ml of acetonitrile were added as protein precipitant. The samples were vortexed again for 1.5 min and then centrifuged at 3000 Hz for 10 min. the clear supernatant was transferred and treated similarly to the previous procedure, except that the microcentrifuge was adjusted to 12,000.Hz for 6 min. (Further more, at the zero time, the supernatant after centrifuging the 200 µl diluted samples and transfer them to a similar eppendrof tube; where we inject from

it 50 µl on the HPLC column, the rest we added to them 50 µl of the WSS, vortexed for 30 seconds and the injected onto the HPLC column for the second time but this time with glibenclamide WSS, this made our detection of glibenclanide peak much easier and more precise.)

During the clinical study serum samples collected on the first Sunday were treated similarly to the samples collected on the next Sunday; moreover, volunteers conditions were the same in the two study days, that is if one volunteer did an exception on the first Sunday he was to do the same on the next Sunday. Glibenclamide serum concentrations were determined by the internal standard method using peak height ratio (glibenclamide / flufenamic acid), such as; if inject 50 µl, this 50 µl represents or include 0.5 µg WISS flufenamic acid, so peak concentration of WISS corresponds to 0.5 µg, thus the peak concentration of glibenclamide will be calculated.

Pharmacokinetic analysis

The maximum glibenclamide serum concentrations (Cmax) and the corresponding peak times (Tmax) were determined by inspection of the individual data and individual drug serum concentration-time profiles. The area under the serum concentration (AUC) time curve was obtained, for each volunteer and for all, by the linear trapezoidal rule.

Statistical analysis

The mean serum levels as well as the standard deviation (SD) of glibenclamide and glucose at each sampling time were calculated for each product. Data were assessed, by the Jandel sigmastat statistical software version 2.0 1995, using the Mann-Whitney rank sum test (a non-parametric comparison), and 95% confidence interval analysis with a minimum level for a significant difference set at $P<0.05$. All data are reported as mean ± standard deviation.

CHAPTER III

RESULTS

In vitro studies

Content uniformity analysis revealed that the two products were within the official compendial limits (95-105 %). The mean glibenclamide content of tablets from brand A was 97.479 % (n=10) compared to 99.966 % (n=10) for brand B. Both products also passed the disintegration test of the BP 98 for plain tablets. The average disintegration times were 1 min 50 sec ± 0.2 min and 50 sec ± 0.03 min for brand A and B respectively , as shown in table 2.

The dissolution of the two products was performed according to the USP XXI in phosphate buffer at pH 7.4 and 37°c. The percent glibenclamide dissolved at predetermined time intervals (10, 30, 60, and 90 minutes) is shown in table 3. About 23.74% of glibenclamide content of brand B dissolved in the first 10 min, compared to 23.31% dissolved for brand A. At 30 min, the percent dissolved were 36.88% and 39.09% for brand A and B tablets respectively. Moreover, at 60 min the percent dissolved was 50.17% for brand B compared to 45.62% dissolved for brand A. At the end of the dissolution study (90min), the percent dissolved for brand B was 58.80% compared to 55.46% dissolved for brand A. Furthermore, no statistically significant difference was found between the percent dissolved at each sampling time between the two brands, as reflected by figure 4.

Pharmacokinetics

Individual pharmacokinetic parameters obtained from the serum concentration-time curve are presented in table 4 and 5. The mean serum concentration versus time curves for the two brands are represented by figure 5. The C-max values for brand A (103.92 ± 43.98 ng/ml) and brand B (98.49 ± 51.26 ng/ml) were not found to be statistically different. Similarly, the T-max values (2.6 ± 0.65 hours and 2.3 ± 0.88 hours for brands A and B, respectively) were not statistically different. In addition, the difference between the AUC_{0-10} for A (380.86 ± 158.18 ng/ml) and AUC_{0-10} for B (359.79 ± 164.98 ng/ml) found in table 6, was not statistically significant ($P > 0.05$ as shown in table 7); using the Mann-Whitney rank sum test.

Pharmacodynamics

The changes in serum glucose concentration versus time are shown in figure 6. Pattern of fluctuation in serum glucose concentration were very similar for the two products. At 3 hours, post drug administration of either products, serum glucose concentrations reached the lowest values (56.7 ± 6.83 mg/dl and 60.6 ± 7.73 mg/dl for Drug A and B respectively), compared with the fasting glucose levels 81.83 ± 9.22 mg/dl and 89.5 ± 8.64 mg/dl for A and B respectively. Between 6 and 8 hours post drug administration, the mean serum glucose concentration started to elevate until reaching 94.2 ± 14.96 mg/dl and 94.0 ± 9.54 mg/dl for brands A and B at 8 hours respectively. Furthermore, at 10 hours post drug administration, glucose serum levels were close to the fasting ones; therefore, statistical analysis of glucose serum concentration at each sampling time did not show any significant differences between the two products.

CHAPTER IV

DISCUSSION & CONCLUSION

The behavioral exhibition shown by brand B regarding content uniformity, disintegration and dissolution were similar to those of brand A; thus, clearly demonstrating that brand B is a pharmaceutical equivalent to brand A with respect to the in vitro results.

The in vivo results, on the other hand, demonstrate that brand B is bioequivalent to brand A in terms of bioavailability and pharmacodynamic effects. Large inter-and intra-individual variation were observed in C-max and T-max following oral administration. For product A, the C-max values ranged between 41.34 -173.6 ng/ml with a mean of 103.92 ± 43.53 ng/ml, whereas the corresponding values for product B ranged between 40.15 -178.86 ng/ml with a mean of 98.49 ± 51.25 ng/ml. The T-max values ranged between 1.5-4 hours for product A, with a mean of 2.6 ± 0.65 hours, as for product B, the T-max values ranged between 1- 4 hours, with a mean of 2.3 ± 0.88 hours. In addition, no significant differences were found in the total AUCs (380.86 ± 158.18 and 359.79 ± 164.97 ng hr/ml for A and B respectively).

Lack of significant differences in AUC values, C-max at T-max between the two products indicate that the two formulations are closely similar in terms of their pharmacokinetic properties. This suggests that the in vivo dissolution and the absorption rate are closely identical for the two products. Furthermore, this in vivo finding is consistent with the in vitro release pattern shown in the dissolution curve .

Both products produced similar effects on blood glucose levels following oral administration of either brand. For both products, the fluctuation in glucose levels

with time seems to reflect the net effect of the drug and food intake. The drastic decline in glucose levels observed between 1.5 and 6 hours post drug administration, is of interest, as it coincide with attainment of peak concentration of glibenclamide; even though there is no clear relation between drug levels and its effect .

The biggest limitation faced by this study was the limited volunteers number, thus implementing a great variation in our SD.

In conclusion, the results obtained from this study demonstrate clearly that the two products of glibenclamide included in this investigation are bioequivalent in terms of both bioavailability and therapeutic efficacy in normal adult individuals.

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Table # 1

Demographic Data										
Volunteers	M.M	C.M	S.H	S.Y	B.S	N.R	R.B	N.B	G.C	A.F
Age (years)	30	26	23	22	26	25	26	25	25	24
Weight (Kg)	77	73	120	86	78	80	75	63	84	90
Height (cm)	188	191	191	170	176	176	172	162	175	170
Race / Sex	W / M	W / M	W / M	W / M	B / M	W / M	W / M	W / M	W / M	W / M
Fasting Glu (mg/dl)	90	86	90	102	85	83	102	90	94	96
Diabetic	No	No	No	No	No	No	No	No	No	No
Renal problems	No	No	No	No	No	No	No	No	No	No
Hepatic problems	No	No	No	No	No	No	No	No	No	No
Cardiac problems	No	No	No	No	No	No	No	No	No	No
Social history	5 cigarette/d 3 coffee cups/d	20 cig/d 3 coffee cups/d	10 cig/d 2 coffee cups/d	No	No	No	1 coffee cup/day	3 coffee cups/day	1coffee cup/day	20 cig/d 1coffee cup/d
Medical history	- Appendix -Inguinal hernia	No	Hyper TG 248mg/dl	Inguinal hernia	No	No	No	No	No	Dyslipidemia Gone with diet
Medication history	No	No	No	No	No	No	No	No	No	No
Blood pressure	115/65	100/60	120/70	125/90	120/70	115/75	125/65	105/70	115/70	105/75
HR / RR	70/12	60/12	64/12	70/12	72/12	72/12	80/12	70/12	75/12	70/12

Table#2. In vitro content uniformity and disintegration

	Obtained values (Daonil)	Obtained values (Glibamid)	Specification value	Reference
Average weight (g)	0.15894 g	0.1615g	0.148 - 0.172	BP 98
Hardness (Kp)	9.3 Kp	6.5 Kp	5 – 2	Internal reference
Thickness (mm)	2.6585 mm	2.8405mm	2.65 –2.95	Internal reference
Friability (%)	0.115 %	0.110 %	Max 1%	USP 23/BP 98
Disintegration (min/sec)	1min 50 sec	50 sec	Max 10 min	USP 23/BP 98
Assay %	97.479%	99.996 %	95 – 105%	BP 98

Table # 3. In vitro – Dissolution result

Time (min)	% Glibamid	% Daonil
10	23.74	22.31
30	36.88	39.09
60	50.17	45.62
90	58.80	55.46

Table # 4. Daonil ®(T-max / C-max / AUC)

Subject	C-max	T-max	AUC
1	173.6	3	476.67
2	70	2.5	249.625
3	65.5	2.5	337.1
4	118	2	307.675
5	138.3	2.5	702.295
6	161.1	1.5	542.1325
7	41.344	4	192.6315
8	75.68	3	357.97
9	113.6	2.5	446.9125
10	82	2.5	295.5675
Mean	**103.9224**	**2.6**	**390.8639**
SD	**43.98424**	**0.658281**	**152.6116**
CI	**27.26102**	**0.40801**	**94.58777**

Table # 5. Glibamid® (T-max / C-max / AUC)

Subjects	C-max	T-max	AUC
1	176.5	3	468.325
2	67.9	2.5	235.025
3	48.7	1.5	310.825
4	65.2	1	273.175
5	143.66	1.5	736.105
6	178.76	2	480.1875
7	40.15	4	257.8625
8	71.64	3	221.17
9	110.326	2	355.2518
10	82	2.5	269.97
Mean	**98.4936**	**2.3**	**360.7897**
SD	**51.25614**	**0.888194**	**160.2158**
CI	**31.76817**	**0.550377**	**99.63345**

Table # 6. Area under the curve for drug A and drug B

subject	AUC- Daonil®	AUC- Glibamid®
1	476.67	468.325
2	249.625	235.025
3	337.1	310.825
4	307.675	273.175
5	702.295	736.075
6	542.1925	480.2175
7	192.6315	257.8625
8	357.97	221.17
9	446.9125	355.2518
10	295.5675	269.97
Mean	**390.8639**	**360.7897**
SD	**152.6116**	**160.2158**
CI	**94.58777**	**99.63345**

Table # 7. Statistical differences between drug A & B at different time intervals

Time (hours)	Statistical Differences
0	NS
0.5	NS
1	NS
1.5	NS
2	NS
2.5	NS
3	NS
4	NS
5	NS
6	NS
8	NS
10	NS

NS: Not Statistically Significant; P > 0.05

Figure 1. WSS + WISS

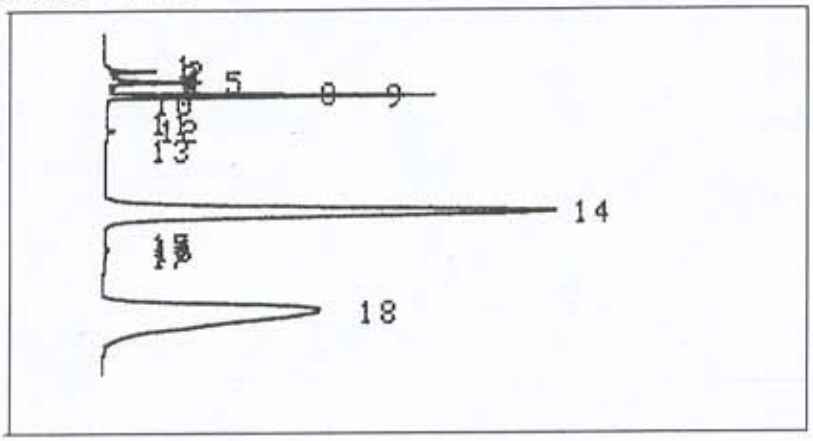


Figure 2. WSS + WISS + SERUM

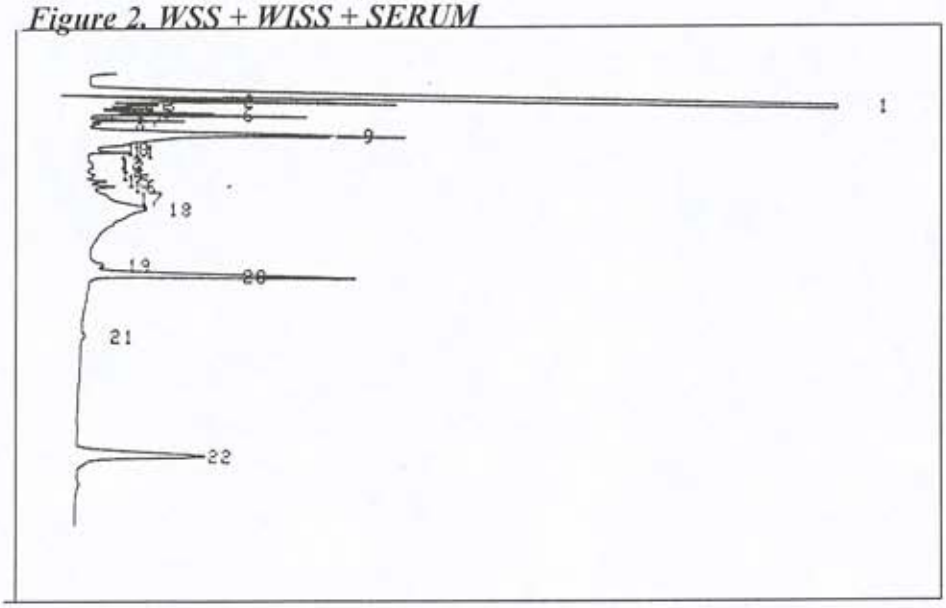


Figure 3. Serum sample at 2.5 hours

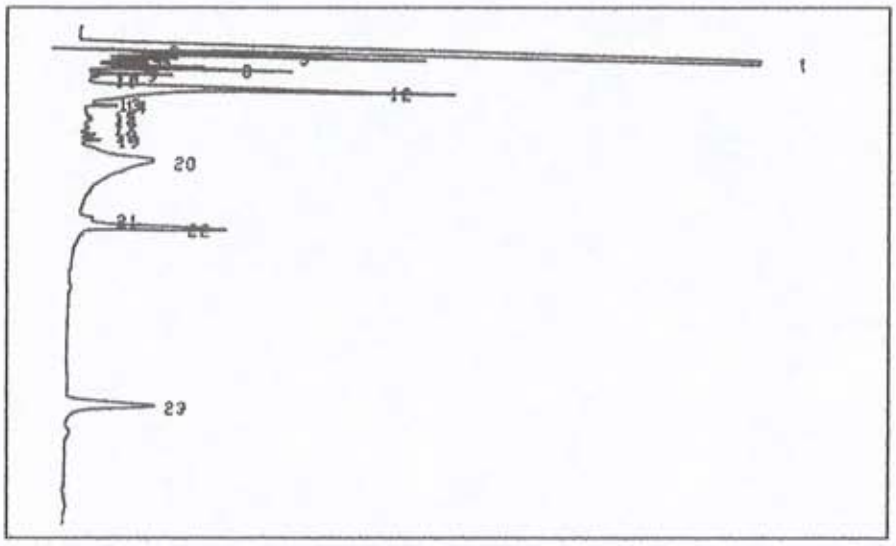


Figure 4. Dissolution curves for drugs A&B

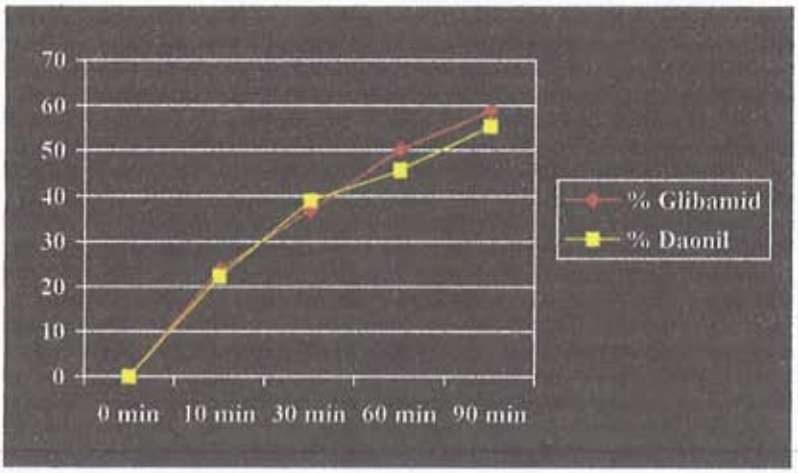


Figure 5. Mean serum concentration vs time for the 2 products

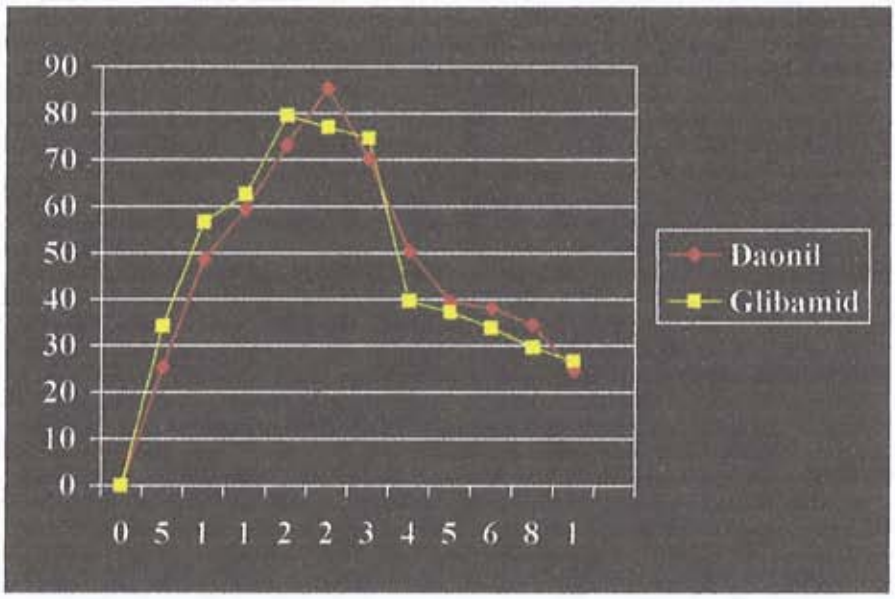
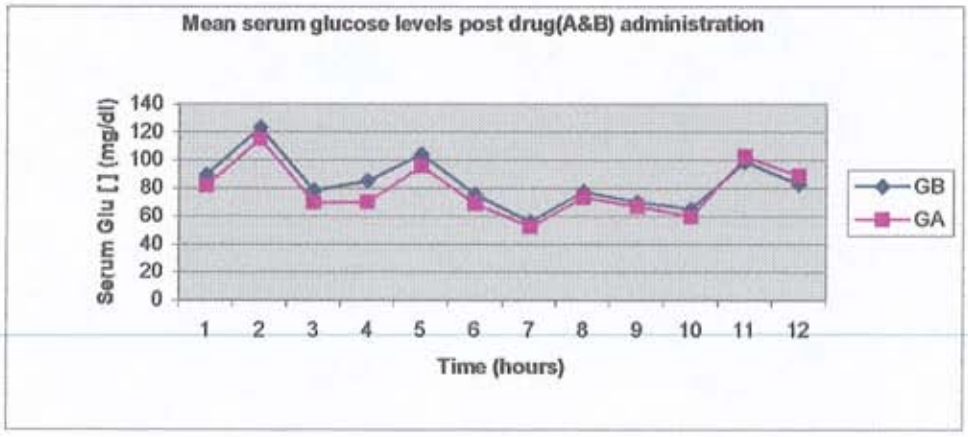


Figure 6. Mean serum glucose levels post dosing



Informed consent form

Study title: A comparative single dose bioequivalence study of two glibenclamide brands among healthy volunteers.

Place : Notre Dame De Secoure Hospital (NDDSH), Byblos.

Investigators : Jean G.Dib, R.ph, pharm.D .
Elias F.Tannous, R.ph, pharm.D candidate.

You will be asked to participate in this research study, if you eligibly fit the physical exam and lab results; so, being a healthy volunteer, makes our principle of medical ethics to inform you about the purpose and benefits of the project, the research methods to be used, the potential risks or hazards of participation and the right to ask for further information at any time during the research procedure. You have the right to know whether medical treatment or compensation is available for physical injuries incurred as a result of participation in the project. Your choice to participate is a voluntary one, and you are free to withdraw from the research at any time. **Your signature at the end of this consent form will indicate that the principle investigator, or his/her agent, has answered all your questions and that you voluntarily consent to participate in this investigation.**

Purpose of this study

This study is to compare the bioavailability and pharmacokinetic properties (absorption distribution metabolism and elimination) of two different glibenclamide formulations (Glibamid® versus Daonil®) in a single dose of 5mg, among healthy volunteers.

Procedures

Your participation in this study is for two consecutive Sundays, where each Sunday you will stay for approximately ten hours, separated by a one week interval.

Prestudy evaluation

If you agree to participate in this study, one week before being in rolled, you will be asked to do a free complete physical exam with some blood, serum and urine analysis, also you will be asked to answer some questions about your health, previous or current hospitalization and medications; moreover, you will be asked to abstain from taking any drugs for at least one week prior to the study.

While on study

You will be transported to Notre Dame De Secours Hospital, by an assigned bus, on two consecutive Sunday mornings after being fasting for 10 hours before the study takes place. Medications (1tablet / day) will be given at 7:30 AM and blood samples will be taken for a period of 10 hours thereafter. The study will be divided into two periods where the two drugs will be orally taken in a randomized order one each Sunday. At 7:15 AM, each volunteer will receive a 200 ml of orange juice with 5mg of sugar; at 7:30 AM, blood sample will be taken and each subject will receive a single oral dose of 5mg of either brands with 200ml of water. Breakfast will be served 1.5 hours after initiation of therapy, a 200ml of orange juice at 4 hours, a standardized lunch at 6 hours, and a light snack at 8 hours. During the study day, you will be abstained from strenuous activities and smoking. Blood samples will be obtained by a registered nurse from the forearm veins (one injection for the whole day). A blood sample (10 ml) will be withdrawn or collected at 0.5,1,1.5,2,2.5,3,4,6,8 and 10 hours

after drug administration. During the study a routine monitoring for your blood glucose level, by a direct glucose test machine, will be performed every half an hour. Entertainment will be offered during the 10 hours stay.

Risks

Glibenclamide , the drug you will take, might produce hypoglycemic effect and gastrointestinal disturbances i.e. nausea, vomiting, diarrhea, anorexia and hunger. These side effects are usually rare and not common. We will try to eliminate them by administering these drugs with food and fluids. In addition, the sponsoring company will insure the volunteers during the study days thus adding additional protection for any inconvenience.

Potential Benefits

1) To the public and patient. This study able us to determine whether Daonil® and Glibamid® are equally efficacious and similar in their pharmacokinetics properties which helps clinicians to utilize the better agent for patients management.

2) For subjects , they will have a CBC, complete physical exam, blood and urine analysis, all free from charge, along with 100,000 L.L for participating. Labeled files containing the above results will be provided to the volunteers as their base line records.

Confidentiality and Anonymity

Information related to you will be treated in strict confidence to the extent provided by law . your identity will be coded and will not be associated with any published results.

Freedom to withdraw

Your participation in this study is voluntary. You may refuse to participate or discontinue your participation during the study at any time without prejudice and without affecting future health care.

Volunteers contact

If you have additional questions about this study, please do not hesitate to contact

Jean G. Dib,Pharm-D mobile :03/408926

Elias F. Tannous,R.Ph mobile :03/503781

I have read and understand this consent form, and I am willing to participate in the study,

Volunteers' name	Volunteers'Signature	Date
Witness's Name (typed or printed)	Witness's signature	Date

I have explained the purpose of this study to the volunteer subject. To the best of my knowledge , s/he understands the purpose, procedure, risks and benefits of this study,

Investigator's Name	Investigator's signature	Date
Witness's Name		*witness's signature*
Date		