

Original Research Article**Comparative Evaluation of the Quality Control Parameters and
Hypoglycemic Drug Effect of Some Brands of Glibenclamide
Tablet****ABSTRACT**

A comparative evaluation of the quality control parameters and hypoglycemic drug performance check of some brands of glibenclamide tablets was studied. The physicochemical parameters of the brands were evaluated through uniformity of weight, friability and hardness tests along with disintegration and dissolution profile in phosphate buffer (pH 8). The hypoglycemic drug performance of the brands was evaluated by twice daily oral dosing of 5mg glibenclamide tablet under controlled evening meal and fasting blood sugar (FBS) determinations. The statistical inferences about the groups' mean fasting blood sugar (MFBS) were compared with a postulated/expected population FBS of 5mmol/L and the hypothesis that there is no difference in the population from which the FBS obtained for each group treatment were drawn was tested. The brands complied with the USP specifications for tablet hardness, weight uniformity, and friability and disintegration tests. There was no significant difference in the chemical content among the brands at CI=95%. The assay gave a chemical content of $92.4\% < x < 102.5\%$. The dissolution profiles in phosphate buffer revealed satisfactory C_{45} and T_{70} of $\leq 70\%$ and 45 min respectively. The MFBS of each brand laid beyond $2 \times SE$ of the overall mean and postulated population MFBS. The method above can be applied as a performance check for different brands of oral hypoglycemic drugs emanating from the possible differences in the production protocols.

26 **Key words:** Diabetes, Glibenclamide, Hypoglycemic effect, Performance check

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31 **1. INTRODUCTION**

32 Glibenclamide is an oral hypoglycemic agent of the sulphonyl urea group indicated
33 for non-insulin dependent diabetes mellitus (NIDDM) [1]. The uses of drugs based on
34 their generic names encourage free choice of drug among the available brands.
35 Competitive pricing should therefore be matched with comparative drug performance
36 and clinical efficacy. For effective cost reduction to enable affordability,
37 manufacturers may choose active ingredients and recipients that lead to fair
38 manufacturing cost which translates to the market cost [2]. The polymorphic form of
39 drugs employed in the production of glibenclamide may characterize their
40 bioavailability profile [3]. Two forms of glibenclamide are available commercially,
41 namely, the glassy form or amorphous and the crystalline [4].

42 In synthetic chemistry, glibenclamide that exists in the crystalline form is convertible
43 to the glassy form by quench-cooling of the melt and cryogenic milling [4, 5]. The
44 solid state properties of the amorphous samples, X-ray powder diffraction (XRD),
45 Infra-red spectroscopy (FTIR), ultra-performance liquid chromatography (UPLC) and
46 broad-band dielectric spectroscopy (BDS) have been exploited to distinguish the two
47 forms based on their physiochemical profiles [4-6].

48 In quality control studies, it has been revealed that some drugs pass the quality
49 control tests but show biological inequivalency [7]. This has been found to be due
50 largely to the issues of polymorphism and choice of the crystalline form of drug used

as the raw material. Since different lattice energies (and entropies) associated with different polymorphs give differences in physical properties such as solubility and dissolution rates, the variations may have impact on the absorption of drug from the dosage form [8-10]. The most critical issue related to drug substance polymorphism is equilibrium solubility which is the concentration of drug dissolved when there is equilibrium between the solid drug substance and solution. Drug dissolution testing is therefore appropriate for drug product evaluation [11].

This study was aimed at evaluating the quality control parameters of the six brands of glibenclamide sold in the market and to assess the biopharmaceutical indices of the drugs.

2. EXPERIMENTALS

Materials and Chemicals

HPLC system with UV detector, DoubleeGee glucometer (Doublee-Gee, China); acetonitrile (HPLC grade), monobasic potassium phosphate, sodium hydroxide and hydrochloric acid and methanol were all analytical grade and obtained from Merck (Germany). Drug samples GL1, GL2, GL3, GL4, GL5 and GL6 were glibenclamide 5mg tablets procured from registered pharmaceutical outfits in Nigeria. Glibenclamide powder was supplied as a gift sample from Sprem Chemicals and research centre, China and employed as reference powder.

2.1 Methods

Buffer preparation

Monobasic potassium phosphate buffer of pH 8.0 was prepared according to established protocols [12].

Standard Solution

75 An accurately weighed amount of pure glibenclamide powder equivalent to 50mg
76 was dissolved in 0.2M sodium hydroxide in a 50ml volumetric flask to form a
77 concentration of 1mg/ml. A series of solutions of glibenclamide of 0.5–20mg/L were
78 prepared from stock solution by diluting accurate volumes of the stock solution in
79 phosphate buffer (pH, 8.0). A calibration curve was prepared using the dilutions.

80 *Calibration Graph*

81 Linear calibration graphs for the determination of glibenclamide (0.5 – 20mg/L) was
82 plotted and the linear–regression analysis expressed in Fig. 2.

83 *Linearity*

84 The calibration curve was analyzed for linearity using the least square regression
85 method with triplicate determinations for each concentration.

86 *Sample preparation*

87 An accurately weighed amount of tablet powder of the six samples equivalent to
88 12.5mg were individually transferred to a 50ml volumetric flask and 20ml of
89 acetonitrile was added and sonicated for 5min. A 5.0ml aliquot of the resulting
90 solution was mixed with the mobile phase to a volume of 1L.

91 *Drug quality control Parameters*

92 *Disintegration Test*

93 The tablet disintegration was determined at 37°C using a Veego model VTDH₃
94 disintegration testing apparatus (Rutartek, India).

95 *Weight Uniformity*

96 Tablets of each brand were weighed individual using a digital analytical balance
97 (Adventure Ohaus) and the mean calculated. The percentage deviation of the
98 individual tablets from the mean was determined.

99 *Tablet hardness Test*

The crushing strength of the tablets was determined using a Monsanto tablet hardness tester at 25 per/min.

Assay/Chemical Content

Twenty tablets of each six of the brands were weighed separately and crushed in a mortar. The powdered drug product of glibenclamide were weighed and an amount equivalent to 5mg was weighed in triplicate into conical flasks and dissolved in methanol to produce 50µg/ml of glibenclamide. The sample was injected into the chromatographic system and analyzed at 229.5nm,

Dissolution test

Dissolution test was performed using an Erweka DT-6 dissolution tester according to dissolution tests protocols. The dissolution medium was 900ml comprising of monobasic potassium phosphate buffer at $37 \pm 0.5^{\circ} \text{C}$ and the speed of rotation 100 rpm. At time 5, 15, 30, 45, 60 and 90 min, 5ml aliquot samples were withdrawn and replaced with equal amounts of fresh medium to maintain a constant volume. The withdrawn samples were analyzed using the HPLC system. . The T_{70} (average time for 70% of the active drug to be released and C_{45} (concentration at 45 minutes) were determined.

Hypoglycemic Performance Evaluation

Thirty six volunteers, oral hypoglycemic users from hospitals in the neighborhood were selected for the study after obtaining their informed consent and administrative approval of the community pharmacy management where the study was undertaken. The inclusion criteria were patients within the age 30 and 60 years, oral hypoglycemic users (Type 2 diabetics) and body weight not less than 50kg. Type 1 diabetics were excluded. The fasting blood sugar (FBS) was taken at 7am on the first day of study and referred to as study entry point (SEP) value. Six volunteers

were given a particular brand of glibenclamide as a twice daily dosing for 6 days and the change in fasting blood sugar (SEP - FBS) values per volunteer every day recorded.

2.2 Statistical Analysis

Statistical significant differences in the values of the change in blood sugar levels for each brand of drug were assessed and analysed using one sample hypothesis and one tailed at $\alpha=0.05$. The difference between the six brands was evaluated using a two sample hypotheses to test for differences in the reductions of fasting blood sugar levels achieved by the various brands. Other parameters were statistically analyzed using SPSS ver. 17 for significant differences at $P<0.05$ [13]

3. RESULT

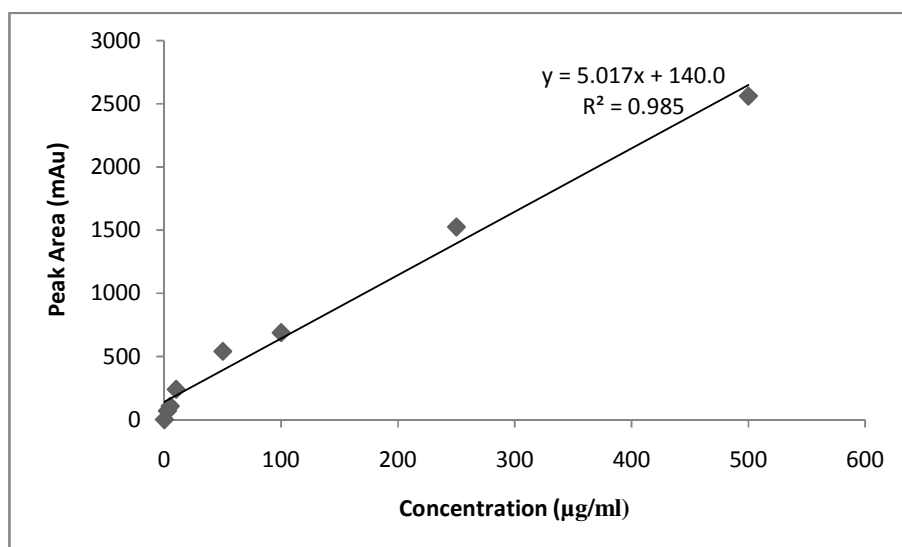
The randomly selected brands of glibenclamide were all within the shelf life and authorized for sale in the country. The brands were also available in the leading drug retail outlets in the study area. There was no preference for one than the other with respect to buyers or prescribers. The volunteers were aged 34.5 ± 2.6 years and body weight 59.4 ± 5.8 kg` with no other known metabolic disease.

Table 1: The details of the drugs used in the study

Name	Code	Manufact Urer	Manufacturing/Expiry date	Nafdac Reg. number	B. No.
Daonil	GL1	May and Baker	08 – 2011/08-2014	04-0744	T028
Glanil	GL2	Nig German chemical	03 – 2011/03-2014	04-2450	W228
Solimide	GL3	Solidum	03 – 2009/02-2012	04-8751	9C-27

		Pharm			
Gliben J	GL4	Juhel	05 – 11/04-2014	04-5735	0019
Clamide	GL5	Hovid	06 – 11/06-2014	04-4015	BB06596
Diabeta	GL6	Greenlife	11 – 10/10-2013	04-3856	DB-02

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144 **Fig. 1: Calibration curve and regression equation of pure glibenclamide**
 145 **powder**

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147 **Table 2: The physicochemical analysis of the brands of glibenclamide in the**
 148 **study.**

Drugs	Tablet Hardness	Weight Uniformity MDM%	Chemical content (%w/w)	Disinte- gration time (min)	Friability (%)	Dissolution profile(SIF)	
						C ₄₅ (%)	T ₇₀ (min)

GL1	3.5±0.3	3.2	98.5±0.2	4.5±0.2	0.03	38	80
GL2	4.4±0.5	1.5	99.4±0.2	4.0±0.2	0.04	36	78
GL3	3.9±0.2	2.4	101.3±0.5	3.0±0.1	0.03	30	83
GL4	4.8±0.8	4.5	105.5±0.6	5.0±0.2	0.02	17	85
GL5	4.8±0.2	2.8	92.4±0.2	4.0±0.2	0	17	83
GL6	4.0±0.4	3.1	99.6±0.5	4.2±0.1	0.01	30	82

Key: MDM- Maximum deviation from the mean

Table 3: The statistical analysis of the hypoglycemic drug effect of the drug products.

Brands	Number of sample	MFBS (Mmol/L)	MSEP (Mmol/L)	Statistical Analysis			
				T _{calc.}	T _{critical} α=0.05	T _{critical} α=0.01	Remark
GL1	105	6.2	7.9	1.65	0.64	1.62	
GL2	111	5.7	8.9	1.97	0.12	1.34	
GL3	106	4.8	7.8	3.41	0.12	1.19	
GL4	94	5.2	10.3	1.19	0.71	1.14	
GL5	101	4.5	9.6	1.94	0.98	1.59	
GL6	89	6.7	8.9	1.49	1..43	1.97	

Key: MFBS- Mean fasting blood sugar; MSEP- Mean Study Entry Point (mmol/L)

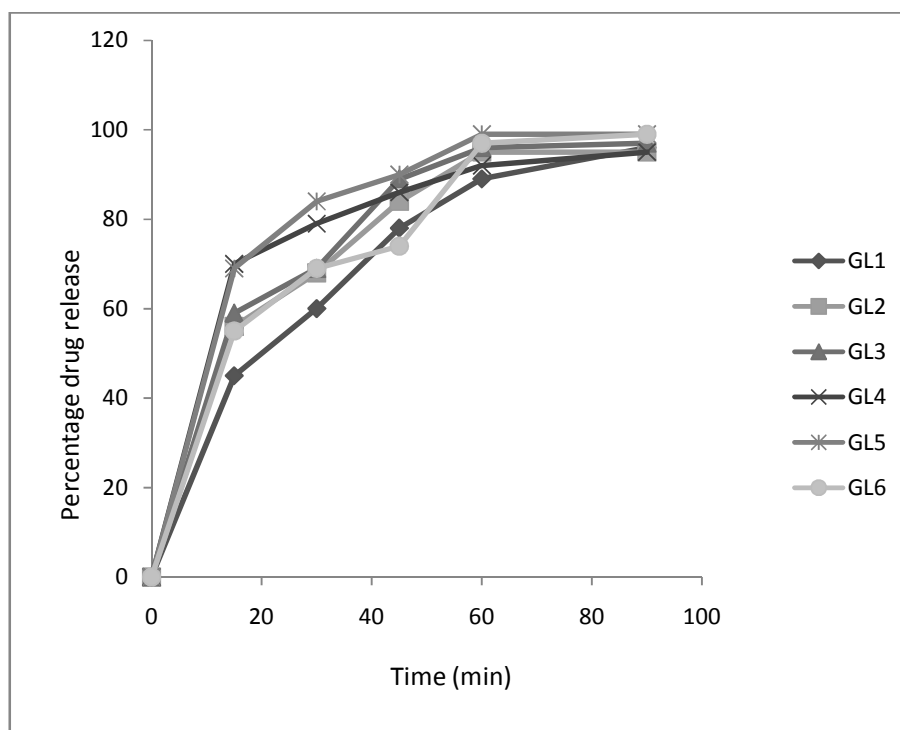


Fig 2: Dissolution profile of the six brands of glibenclamide drug products in dissolution medium (pH 8)

4. DISCUSSION

The drug glibenclamide has been advocated for effective control of blood sugar in Type 2 diabetes. Some physicians present the notion that twice daily dosing of glibenclamide together with controlled evening meal adequately regulates FBS. Upon the increasing number of generic products of the drug, there was the need to assess the quality and performance of some of the products. The brands employed were all registered products (Table 1) in the country and they all complied with the quality control parameters investigated. The widespread peddling of substandard drugs makes the investigation of the quality control parameters expedient. Performances of drugs have been demonstrated to be closely associated with the quality indices of drugs [14]. Glibenclamide is a BCS class 2 drug with low aqueous solubility. Manufacturing protocols from different producers are expected to

circumvent this problem and these may be responsible for the variations in the dissolution characteristics. The dissolution outcome however did not show marked variations as to suggest an indictment on the performance of the various brands. Buffer of pH 8 was used to assess the dissolution characteristics of the brand as this gives the optimum dissolution properties with respect to the acidic nature of the drug and the pH nature of the intestine where maximum absorption is expected. Since drug dissolution affects the bioavailability of the brands the outcome of the dissolution assessment will predict the in-vitro performance. The volunteers willingly accepted their allotted medications and complied with the routine of reporting for FBS checks to the extent of over 90% responsiveness. This was owed to the fact that the enlightenment on the need to achieve a stable FBS was well understood by the volunteers. The expected FBS projected for the group was 5.0. This value was designed as a FBS ideal for effective drug performance. One sample hypothesis and two tail statistical analyses for the observed MFBS in each group with data collection over 21 days determination was analysed to assess if there was difference in the MFBS of the groups compared with the targeted FBS. There was no significant difference in the MFBS of the individual group with the projected FBS at $P < 0.05$ and 0.01. The drugs therefore at the dosing and the feeding condition will not produce a blood pressure of about 5mmol/L [15, 16]. There was however a significant reduction in MFBS when paired T-test was used to test the MSEP and the group MFBS looking at the values of fasting blood sugar for the six brands.

5. CONCLUSION

The study revealed that the various brands of glibenclamide marketed in the country from different sources have comparable quality characteristics. The expected MFBS akin to convenient fasting blood sugar reduction was however not met by any of the

brands which is an indication that a supportive therapy may be needed to achieve this projected value. Failure to meet the projected value may also not be drug or drug production factor.

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