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# Development and Evaluation of Combined Gliclazide and Enalapril Maleate Immediate Release Tablet

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## Abstract

Chronic diseases such as diabetes mellitus and systemic hypertension have high prevalence all over the world. In majority of cases, patients having diabetes mellitus are also suffering from systemic hypertension and vice versa. A**ims:** The purpose of the study is to prepare a combination dosage form which can be used to treat both the diseases concomitantly, reducing pill burden and increasing patient compliance. **Methods:** Based on positive results of a feasibility study, including doctors' opinion and prescription survey, Gliclazide and Enalapril maleate were selected as the active ingredients for developing a FDC (Fixed Dose Combination) preparation. Immediate release combination tablet (Gliclazide and Enalapril maleate Tablet, GET), containing 80 mg of Gliclazide and 5 mg of Enalapril maleate, were prepared by direct compression method having diameter of 8.7 mm and thickness of 3.45 mm. Their physical properties were determined. The tablets were subjected to dissolution testing in a six-station USP paddle apparatus rotated at 100 rpm. The dissolution medium was pH 7.4 phosphate buffer maintained at 37$^{o}$C. The prepared tablets were again subjected to stability testing at conditions of 25°C/60%RH and 40°C/75%RH for 6 months. Shelf-lives of the drugs were determined following ICH guidelines as well as by kinetic calculations. **Results:** The prepared tablets showed hardness of 6.13 kg.cm$^2$ and friability value of 0.1676%. The dissolution of Gliclazide and Enalapril maleate was 98% and 85% respectively at 45 minutes. During stability testing, slight discoloration of the tablets was observed at higher temperature although the assay results were found to be satisfactory. **Conclusion:** The prepared tablets met the pharmacopeial standards.

**Keywords:** Gliclazide, Enalapril maleate, combined tablet dosages form, fixed dose combination, dissolution studies.

## INTRODUCTION

There is demonstrable evidence to show that the incidence of diabetes mellitus is rising rapidly in population of the developing countries of Africa and Asia [1]. International Diabetes Federation gives high estimates of occurrence of diabetes in Bangladesh, Pakistan, Sri Lanka, Nepal and other countries of the region [1, 2] .

Patients with diabetes mellitus have an increased prevalence of hypertension and associated cardiovascular disease (CVD). The risk of an individual of developing CVD is much greater when both diseases coexist [3]. Although population statistics of diabetic-antihypertensive patients in Bangladesh are not available, a survey on the slum dwellers of Dhaka city revealed that 7 out of 1000 adults are suffering from diabetes and hypertension [4]. An international investigation revealed that hypertension affects about 60% of patients with type II diabetes [5]. Thus, concomitant antihypertensive therapy is frequently required along with diabetes treatment. Our recent study on the prescribing pattern of diabetic-hypertensive patients revealed that out of the 534 prescriptions, 225 contained both oral antidiabetic and antihypertensive drugs [6]. Studies have shown that the use of angiotensin converting enzyme (ACE) inhibitors can prevent the progression of renal damage and delay progression to end-stage renal disease in addition to lowering blood pressure [5, 7, 8]. In Bangladesh, the physicians are prescribing the ACE inhibitors at a greater frequency in diabetic-hypertensive patients and the same trend is observed in other countries [5, 6, 7, 8]. Such patients are exposed to pill burden having compelled to take a large number of dosage forms for treat of their complicated disease conditions.

A fixed dose combination (FDC) is a formulation of two or more active ingredients combined in a single dosage form and available in certain fixed doses [9]. FDC pharmaceutical products can be used to treat the same disease state, multiple disease states or counteract the negative side-effects [10]. The obvious strengths of fixed-dose drug combinations include the potential advantages of increased compliance, convenience and cost savings. In contrast, potential disadvantages include reduced flexibility in dosing, exposure of some patients to therapies they do not require and possible increased risks of

adverse effects without added benefits [10, 11].

In recent days, the pharmaceutical industry is placing a greater emphasis on FDC products. There are lots of combination drug products in the consumer market, available both locally and globally [10, 12]. Recent inclusions are Caduet (Pfizer), Vytorin (Merck/Schering-Plough), Truvada (Gliead Sciences), Symbax (Eli Lilly and Company), Lotrel (Novartis) etc.; many of which are approved by FDA and other concerned authorities [10, 13]. Caduet is unique in being the first product that combines two drugs (amlodipine besylate and atorvastatin calcium) to treat two different, but concomitant disease states.

Considering the prevalence of diabetic-hypertension as well as the prescribing pattern of drugs in patients suffering from such complications, it is apparent that antidiabetic and antihypertensive drugs seem to be potential candidates for incorporating in a FDC product. Outcome of a feasibility study of developing an antidiabetic-antihypertensive combination dosage form, which included survey of prescribing pattern and doctors' opinion, further rationalized the suitability to formulate such a combination product containing an oral hypoglycemic agent and an ACE inhibitor [6]. The prescription survey revealed that among 157 patients receiving Gliclazide, 149 of them concomitantly received an ACE inhibitor (Enalapril, ramipril or captopril) [6]. In this context, an immediate release combination tablet containing Gliclazide and Enalapril maleate was developed and evaluated for various pharmaceutic properties including dissolution and stability testing.

## MATERIALS AND METHODS

**Materials.** Gliclazide BP (Shandong Keyuan Pharmaceutical Co. Ltd., China, potency 99.6%) and Enalapril maleate USP (Neuland Laboratories Limited, India, potency 99.7%) were kind gifts from Orion Laboratories Ltd. and Eskayef Bangladesh Ltd. respectively. All other ingredients were of pharmaceutical grade. Chemicals used for analytical and dissolution studies were of reagent grade.

**Methods.**

**Formulation and preparation of combined Gliclazide and Enalapril maleate tablet (GET).** Formulation and preparation of Gliclazide 80mg tablet was established in our laboratory in a previous study [14]. For the present work, a modified formula was adopted by trial and error in which Enalapril maleate was incorporated along with Gliclazide. The formulation of the GET is shown in Table 1. The tablets were prepared by direct compression method using a single punch tablet machine (Erweka AMD, Germany) after proper blending of the ingredients and 2kN pressure was applied. The blending was done on a laboratory scale Double Cone Blender (Model No.:C – 227, Company name: Solace Engineering Corporation, India) for an adequate time period. We have checked the content uniformity of the tablets to ensure proper blending. Two hundred tablets were prepared having diameter of 8.7 mm and thickness of 3.45 mm.

**Table 1:** Formulation of Gliclazide and Enalapril maleate immediate release tablets (GET).

| ***Ingredients*** | ***Quantity per tablet (mg)***[a] | ***Purpose*** |
|---|---|---|
| Gliclazide BP | 80.25[b] | Hypoglycemic |
| Enalapril maleate USP | 05.02[b] | Antihypertensive |
| Microcrystalline cellulose BP (Avicel PH103) | 60.00 | Disintegrant |
| Lactose BP | 48.00 | Filler |
| Sodium starch glycolate BP | 03.00 | Disintegrant |
| Magnesium stearate BP | 01.50 | Lubricant |
| Colloidal silica BP (Aerosil 200) | 03.00 | Glidant |

**[a] total weight of the tablet is 200.77 mg.**
**[b] calculated on the basis of raw material potency.**

**Dissolution studies of GET.** Dissolution studies were conducted on an USP type II apparatus having a six paddle assembly (model Pharma Test PT-DT7, Germany)

The procedure for following Gliclazide dissolution was same as stated in BP 2007 except that a multipoint analysis was carried out instead of a single point determination in order to obtain the full drug release pattern [15]. The dissolution medium was 900 ml phosphate buffer of pH 7.4 maintained at a temperature of 37±1ºC and the stirring rate was maintained at 100 rpm. Six tablets were tested in each case. Samples were withdrawn at every 5 minutes time intervals up to 45 minutes and their absorbance values were recorded at 226 nm and 290 nm after filtration and suitable dilution as mentioned in the BP 2007 [15]. The absorbance obtained at 226 nm was corrected by subtracting the absorbance obtained at 290 nm (according to the official procedure stated in BP 2007). The amounts of drug present in the samples were calculated with the help of a calibration curve constructed from standard solutions of different concentration of Gliclazide in the dissolution medium. For determining Enalapril maleate dissolution, the same procedure was followed as mentioned for Gliclazide, which is a minor modification of the method developed by Lokesh and Naidu (2007) and is widely used in the industry for its simplicity [16]. USP 2007 also states a phosphate buffer of pH 6.8 for testing dissolution of Enalapril maleate tablets [17]. However, absorbance of the samples was recorded at 210 nm and Enalapril maleate concentration was determined using a suitable calibration curve. The Beer-Lambert's law was found to be obeyed between the concentration range of 3 µg/ml to 30 µg/ml in pH 7.4 phosphate buffer. The analysis of the drugs were also verified by randomly cross-checking some of the samples by simultaneous UV spectrophotometric equation method developed for the assay of the active ingredients in the prepared tablets which will be reported elsewhere [6, 18].

**Other tests**. General appearance, disintegration time, hardness, friability, weight variation, content uniformity and loss on drying were determined according to the standard procedures [15, 19]. Assay for the active ingredients was done by a newly developed simultaneous UV spectrophotometric equation method (Vierodt's method) which will be reported elsewhere [6, 18] .

**Stability testing of GET.** The prepared tablets were blister packed using aluminium-PVC foil, kept in stability testing chambers and subjected to stress conditions of 40ºC, 75%RH in addition to those kept at 25ºC, 60%RH. These conditions were chosen according to the guidelines set forth by ICH (International Conference on Harmonisation) [20]. Samples were stored for six months and assay for the active ingredients in the tablets were conducted in the middle (90 days) and at the end of the test period (180 days) by simultaneous UV spectrophotometric equation method (Vierodt's method). Appearance, average weight, hardness, friability and loss on drying of the tablets were also determined at the stated time intervals.

## RESULTS AND DISCUSSION

Immediate release GET tablets were formulated and prepared having strengths of Gliclazide 80 mg and Enalapril maleate 5 mg per tablet by direct compression. Although various technologies are becoming available for incorporating two or more drugs in the same dosage form, direct compression method to prepare a tablet dosage form is likely to be the most versatile and economic way avoiding complicated procedures and expensive machineries [10]. However, it should be ensured that the active ingredients are compatible with each other and with the excipients in the formulation.

The general appearance of GET was found satisfactory having a white color and no markable odor. Friability was found to be 0.1676% with a hardness of 6.13 kg/cm$^2$ indicating that the tablets were of sufficient mechanical strength. Weight variation of GET was within the range of –0.08% and +0.15% of the average weight (201.85 mg) and complied with the BP specification (accepetd range ±7.5%). Content uniformity for both glicazide and Enalapril maleate in the tablets also satisfied the compendial requirements.

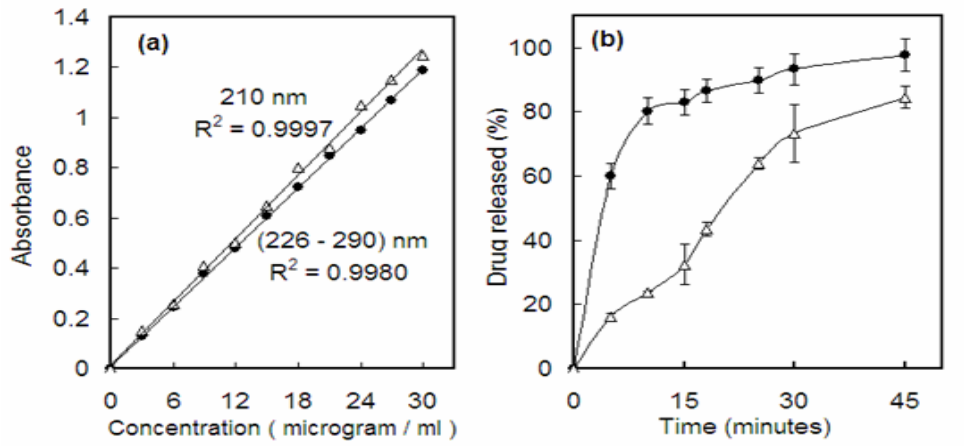


**Figure 1**. (a) Calibration curves for Gliclazide and Enalapril maleate in pH 7.4 phosphate buffer medium. (b) Dissolution profiles of GET (mean ± std. dev., n = 6) in pH 7.4 phosphate buffer medium. Symbols: • Gliclazide, Δ Enalapril maleate.

The disintegration time for GET was found to be 1.5±0.3 minutes indicating a satisfactory value for an immediate release tablet (BP allows 15 minutes for such tablets) [15].

**Dissolution of Gliclazide and Enalapril maleate from GET.** Dissolution of the two drugs was monitored in a medium of pH 7.4 phosphate buffer at 100 rpm stirring speed for 45 minutes. Figure 1a represents the calibration curves of the test drugs. It is evident that the Beer-Lambert's law is well obeyed within the stated concentration range for both Gliclazide and Enalapril maleate. Dissolution profiles, depicted in Figure 1b, are indicative of a much faster release of Gliclazide in buffer medium of pH 7.4 in comparison to Enalapril maleate. This may be attributed to slightly decreased solubility of this antihypertensive in pH 7.4 medium (maximum at pH 7.2) [16]. Dissolution of Gliclazide after 20, 30 and 45 minutes were 89.16%, 95.52% and 98.76% respectively whereas Enalapril maleate dissolved to the extents of 21.85%, 73.82% and 88.25% at the same time intervals. Thus, dissolution of GET met the compendial requirement of minimum dissolution as more than 80.0% of both the drugs were released within the specified time limit of 45 minutes [15]. The liberation of Gliclazide and Enalapril maleate from the combined tablet dosage form in adequate quantities is an indirect indication of the absence of any interference between theses two drugs. However, such statement can only be confirmed by additional studies such as DSC.

**Stability testing of GET.** It is not feasible to investigate the complete stability profile before finalizing a dosage form along with its intended container-closure system. However, the prepared tablets were subjected to 25°C, 60%RH and 40°C, 75%RH, as per guidelines of the ICH [20]. This was done in order to get some preliminary stability information about GET so that necessary modifications in the formulation, processing and packaging can be undertaken if necessary. The information obtained was analyzed as suggested by ICH, the results of which are shown in Figure 2 [21].

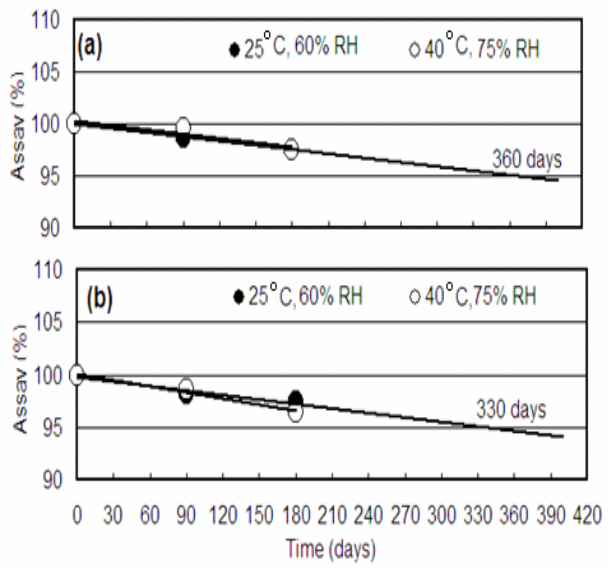


**Figure 2.** Shelf-life estimation of (a) Gliclazide and (b) Enalapril maleate by trend-line extrapolation based on assay of the test drugs as per ICH guidelines. Acceptance level is ±5% according to BP and ±10% according to USP.

As per BP specification, both drugs must be present in the dosage form within ±5% of the label claim (95 mg to 105 mg for Gliclazide and 4.75 mg to 5.25 mg for Enalapril maleate) [15]. From Figure 2a and 2b, it was evident that both the drugs suffered degradation during the experimental time period of 6 months (180 days). The similar slopes of the stability trend-lines revealed that the degradation rates, particularly for Gliclazide, were not much influenced by the stress condition (40°C, 75% RH) of the experiment in comparison to the normal ambient condition (25°C, 60% RH).

**Determination of rate constant and shelf-life** As suggested by ICH for shelf-life determination, extrapolation of the trend-line for ambient condition (25°C, 60% RH) gave a shelf-life of 360 days and 330 days for Gliclazide and Enalapril maleate respectively, which are not good values in terms of a commercial solid dosage form like tablets. Calculation of the degradation rate constants assuming first-order kinetics and derivation of the shelf-lives for the two drugs were also done by the method stated by Sinko [22]. The two methods gave similar results as shown in Table 2. The estimated shelf life of a drug product is highly influenced by the variability of the measured data [23]. For instance, if USP standards are followed, which allows a deviation of ±10% of the label claim, the calculated or graphically estimated shelf-lives of Gliclazide and Enalapril maleate in the prepared tablets will be more than 730 days and 675 days respectively [17].

The changes observed in the physical parameters during six months study, namely average weight, hardness, friability and loss on drying, were not much significant and remained within the acceptable limits (Table 3). However, a slight color change (from white to off-white) was observed at the applied stress condition of 40°C/75% RH after the stability test period.

## CONCLUSION

Gliclazide is widely used in the treatment of type II Diabetes mellitus and ACE inhibitors have proven to be very useful for the treatment of hypertension. Enalapril is an oral prodrug that is converted by hydrolysis to the active molecule, Enalaprilat. These two drugs are prescribed frequently together to the diabetic-hypertensive patients. After positive results from a feasibility study,

**Table 2:** Shelf-life of Gliclazide and Enalapril maleate in GET derived from 1st order rate calculation and from stability trend-line extrapolation.

| *Drug* | *Initial conc. $C_{100\%}$ (mg)* | *Final conc. $C_t$(mg)* | *Time period t(days)* | *Calculated rate constant $k^c$($day^{-1}$)* | *Calculated shelf-life $t_{95\%}$ (days)$^d$* | *Shelf-life from ICH method (days)$^f$* |
|---|---|---|---|---|---|---|
| Gliclazide | 79.95 | 78.00 | 180 | $1.37 \times 10^{-4}$ | 373 | 360 |
| Enalapril maleate | 4.98 | 4.85 | 100 | $1.46 \times 10^{-4}$ | 349 | 330 |

[c] $k = (2.303 / t) \log (C_{100\%} / C_t)$

[d] $t_{95\%} = (2.303 / k) \log (C_{100\%} / C_{95\%})$, $C_{95\%}$ = conc. after 5% degradation.

[f] see Figure 2 and text for explanation.

**Table 3.** Change of physical parameters of GET during stability testing.

| *Parameter* | *Initial* | *After 6 months* | |
|---|---|---|---|
| | | *25°C,60% RH* | *40°C,75% RH* |
| Color | White | White | Off white |
| Average weight (mg) | 200.8 | 203.5 | 201.0 |
| Hardness (kg/cm$^2$) | 6.13 | 6.01 | 5.71 |
| Friability (%) | 0.1676 | 0.813 | 0.813 |
| Loss on drying (%) | 2.10 | 1.71 | 1.52 |

preliminary development of a FDC of GET was undertaken using the lowest doses prescribed for Gliclazide and Enalapril maleate. The prepared tablets met the pharmacopeial standards although there seems to be some problems regarding stability. Thus, the formulation and processing variables need to be fine-tuned in order to improve the quality and stability of the tablets. A sustained release GET may also be prepared by modifying the formulation which may be helpful to patients taking several doses in a day. It is obvious that the patients would derive certain benefits by the use of this FDC product but the limitations should also be kept in mind to get the maximum advantages as discussed previously. The incompatibility of the active ingredients must be verified by DSC and other appropriate methods. Additionally, a thorough study is required to rationalize and prove the clinical efficacy and safety of such a combined dosage form of antidiabetic-antihypertensive drugs before approving it for patients use.

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## REFERENCES

[1] Ghaffar, A., Reddy, K.S. and Singhi, M. Burden of non-communicable diseases in South Asia. *BMJ*. 2004, *328*, 807-810.

[2] Abu, S.M., Ali, L., Hussain, M.Z., Rumi, M.A., Banu, A, and Khan, A.K.A. Effect of socioeconomic risk factors in prevalence of diabetes between rural and urban populations in Bangladesh. *Diabetes Care.* 1997, *20*, 551-555.

[3] Guzmán, C.B. and Sowers, J.R. Special considerations in the therapy of diabetic hypertension. *Prog. Cardiovasc. Dis.* 1999, *41(6)*, 461-470.

[4] Alam, K.S., Huda, M.N. and Rashid, H.U. Prevalence of diabetes mellitus, hypertension and proteinuria in adult unban slum dwellers. The 52nd Congress of the Japanese Society for Dialysis Therapy 2007. Poster no. IS-P-05.

[5] Johnson, M.L. and Singh, H. Patterns of antihypertensive therapy among patients with diabetes. *J. Gen. Intern. Med.* 2005, *20*, 842–846.

[6] Haque, Afroza. Feasibility study of formulation and preparation of combined Gliclazide and Enalapril maleate immediate release tablet. M. Pharm thesis 2008, submitted to Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Dhaka.

[7] Thamer, M., Ray, N.F. and Taylor, T. Association between antihypertensive drug use and hypoglycemia: a case-control study of diabetic users of insulin or sulfonylureas. *Clin. Ther*. 1999, *21(8)*, 1387-1400.

[8] Shorr, R.I., Ray, W.A., Daugherty, J.R. and Griffin, M.R. Antihypertensives and the risk of serious hypoglycemia in older persons using insulin or sulfonylureas. *JAMA.* 1997, *278(1)*, 40-43.

[9] Wikipedia - the free encyclopedia. web: http://en.wikipedia.org/wiki/ Fixed_dose_combination.

[10] Miller, F.H. Combination products: Industry strategies and innovation takes combination products to new levels. In: Technology Overviews, Drug Delivery Report Autumn/Winter, 2005. pp. 42-46.

[11] Hennekens, C.H. Fixed-dose combination therapy with statins: strengths, limitations, and clinical and regulatory considertions. *Am. J. Cardiovasc. Drugs* 2008, *8(3)*, 155-160.

[12] Chakraborti, A. Fixed dose combinations in therapy. Express Pharma 2007; 1-15 August.

[13] DDDs for combined products. WHO collaborating centre for drug statistics methodology. Norwegian Institute of Public Health, Norway 2008.

[14] Begum, Nazma. Studies on dissolution and some other pharmaceutic parameters of commercially available marketed sample and laboratory made Gliclazide tablets. M. Pharm thesis 2007, submitted to Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Dhaka.

[15] BP. British Pharmacopoeia Commission. The Stationery Office, London. 2007. Volume III (Formulated preparations), pp. 2618-2619, Volume IV (Appendices), pp. A274-A288, A304-A305, A397.

[16] Lokesh, B.V.S. and Naidu, S.R. New dissolution method for the evaluation of Enalapril maleate tablets using pH 7.2 phosphate buffer in vitro and determination of its content by validated UV spectrophotometric method. *JASA.* 2007, *2*, 34-37.

[17] USP. The United States Pharmacopeial Convention, Rockville, MD 20852. 2007. Volume 2, pp. 2030-2031.

[18] Beckett, A.H. and Stenlake, J.B. Practical Pharmaceutical Chemistry. CBS India, 1988, pp. 296-303.

[19] Lachman, L., Lieberman, H.A. and Kanig, J.L. The Theory and Practice of Industrial

Pharmacy. Philadelphia.: Lea & Febiger, 1986, pp. 284-286.

[20] Stability testing of new drug substances and products, Q1A(R2). ICH Harmonised Tripartite Guideline 2003.

[21] Evaluation for stability data, Q1E. ICH Harmonised Tripartite Guideline 2003.

[22] Sinko, P.J. Martin's Physical Pharmacy and Pharmaceutical Sciences. Philadelphia: Lippincott Williams & Wilkins, 2006, pp. 401-402.

[23] Komka, K and Kemeny, S. Effect of the uncertainty of the stability data on the shelf life estimation of pharmaceutical products. *Periodica Polytechnica Ser. Chem. Eng.* 2004, *48(1)*: 41–52.