Clinica Chimica Acta 425 (2013) 54–61

Contents lists available at SciVerse ScienceDirect

Clinica Chimica Acta

journal homepage: www.elsevier.com/locate/clinchim

RP-LC simultaneous quantitation of co-administered drugs for (non-insulin dependent) diabetic mellitus induced dyslipidemia in active pharmaceutical ingredient, pharmaceutical formulations and human serum with UV-detector

Muhammad Saeed Arayne [a], Najma Sultana [b], Arman Tabassum [a,*]

[a] Department of Chemistry, University of Karachi, Karachi 75270, Pakistan
[b] Research Institute of Pharmaceutical Sciences, Faculty of Pharmacy, University of Karachi, Karachi 75270, Pakistan

ARTICLE INFO

Article history:
Received 22 February 2013
Received in revised form 9 May 2013
Accepted 20 June 2013
Available online 7 July 2013

Keywords:
Reversed phase liquid chromatography
Rosuvastatin
Metformin
Glimepiride
Gliquidone

ABSTRACT

Background: Rapid, efficient and accurate RP-HPLC-UV method for the simultaneous determination and quality control of active pharmaceutical ingredient (API), pharmaceutical formulations and human serum containing drugs as rosuvastatin together with metformin, glimepiride and gliquidone has been proposed.
Methods: The chromatographic system comprised mobile phase of methanol:water 90:10 v/v; pH adjusted to 3.0 with *o*-phosphoric acid, at 1 ml/min through Prepacked Purospher Star C_{18} (5 μm, 25 × 0.46 cm) column with UV detection at isosbestic point 231 nm.
Results: The method showed good linearity in the range 0.25–25 μg/ml for metformin and 0.5–50 μg/ml for rosuvastatin, glimepiride and gliquidone with correlation co-efficient ≥ 0.998; (precision %RSD < 2) for all drugs in API, formulations and human serum. The recovery of all drugs was 98.9–101.91% in API and formulations and 99.92–102.08% in human serum.
The sensitivity of method increased when drugs were analyzed after programming the detector at their individual λ_{max} where their LODs shifted down to 5, 3, 10 and 9 ng/ml from 10, 17, 15 and 14 ng/ml when calculated at their isosbestic point respectively at least concentration 0.125 μg/ml for metformin and 0.25 μg/ml for rosuvastatin, glimepiride and gliquidone with correlation co-efficient ≥ 0.998 in each case.
Conclusions: The proposed drugs can be analyzed by this method for routine analysis and clinical studies with sensitivity at nanoscale with small sample volume.

1. Introduction

Diabetes mellitus induces malfunctioning of major organs of the body including cholesterol biosynthesis and causes dyslipidemia. Rosuvastatin (ROS) (Fig. 1) is the agent that prevents from dyslipidemia, hypercholesterolemia induced heart attacks and arteriosclerotic vascular disease [1,2] by reducing low-density-lipoproteins (LDL) and total cholesterol; and increasing high-density-lipoprotein cholesterol [3]. For many patients with non-insulin dependent diabetes mellitus (NIDDM), monotherapy with an oral antidiabetic agent is not sufficient to reach target glycemic goals and multiple drugs may be necessary to achieve adequate control [4]. In such cases combination of metformin (MET) and sulfonylureas is used [5] that lowers the blood glucose level by suppressing hepatic glucose output and enhancing peripheral glucose uptake. Therapeutic drug monitoring becomes necessary for their study in plasma for their pharmacokinetics [6].

Literature survey reveals a number of development methods for quantitation of these drugs. ROS has been determined by UV–visible spectrophotometry [7], tandem-mass spectrometry [8], in biological fluids [9–13] using chromatography [13], chromatography with tandem-mass spectrometry [14,15] and capillary zone electrophoresis [16]. Simultaneous methods have been developed for determination of MET with glyburide in human plasma [17], sulfonulureas [18], glipizide, gliclazide, glibenclamide or glimepiride (GLM) in plasma [19]; glibenclamide from their combined dosage forms [20] and GLM with rifampicin for their pharmacokinetic studies [21].

Our research group has long been working on the development of RP-HPLC methods of individual drugs as well as simultaneous determination of a combination of co-administered drugs in pharmaceutical formulations and human serum as ROS with pioglitazone, gliquidone (GLQ), and simvastatin [22]; with simvastatin, atorvastatin, pravastatin and ceftriaxone [23]; with diltiazem, atorvastatin and simvastatin [24];

Abbreviations: ROS, rosuvastatin; MET, metformin; GLM, glimiperide; GLQ, gliquidone; ACN, acetonitrile; RP-HPLC, reverse phase high performance liquid chromatography; API, Active Pharmaceutical ingredient; ICH, International Conference on the Harmonization of technical requirements for the registration of pharmaceuticals for human use; NIDDM, non-insulin dependent diabetes mellitus.

* Corresponding author.
E-mail addresses: arman_tabassum@hotmail.com, armantabassum@yahoo.com (A. Tabassum).

0009-8981/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.cca.2013.06.020

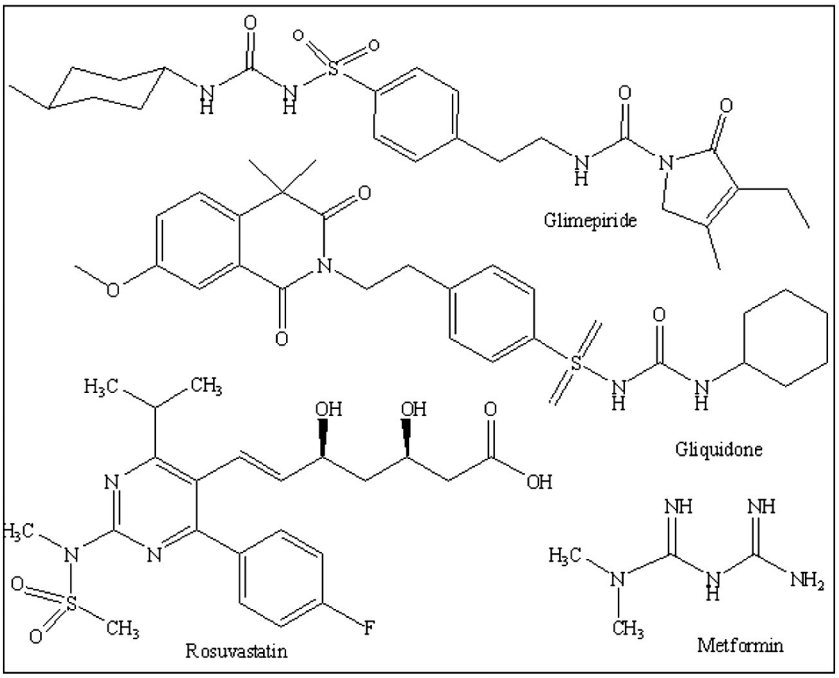

Fig. 1. Chemical structures of drugs.

with lisinopril, pravastatin and atorvastatin [25]; atorvastatin with celecoxib [26]; with ACE inhibitors [27], with atenolol, spironolactone glibenclamide and naproxen [28]; GLQ alone [29]; and with pioglitazone hydrochloride, and verapamil [30]; MET with NSAIDs [31]; and glipizide and GLM by RP-HPLC in dosage formulations and in human serum [32] have been carried out.

In our present work we proposed a method for determination of ROS, MET, GLM and GLQ simultaneously by liquid chromatography without using toxic organic solvents. The results and sensitivity of the method and efficiency of conventional HPLC system magnify with programming the UV detector of the system. The method is economical and can be applied where limited resources are available for the detection and quantitation of analytes present in nanograms in API, formulations and human serum. The proposed method can be a precursor to pharmacokinetic studies of the dosage forms. The method follows ICH (International Conference on the Harmonization of technical requirements for the registration of pharmaceuticals for human use) guidelines [33].

2. Materials and methods

2.1. Materials and reagents

MET (Merck Pvt. Ltd.), ROS (Pharm Evo Pvt. Ltd), GLM (Pharm Evo Pvt. Ltd) and GLQ (Pharmatec Ltd) were used as reference standards without further purification. Glucophage® 250 mg, X-plended® 20 mg, Evopride® 10 mg and Glurenor® 20 mg (the tablet formulations of all API respectively) were purchased from local pharmacy. Serum from a healthy person was collected at Fatmid Foundation Karachi. HPLC grade methanol, acetonitrile (ACN) and *o*-phosphoric acid (85% w/w) were purchased from Merck, Darmstadt, Germany. Double distilled de-ionized water was used throughout.

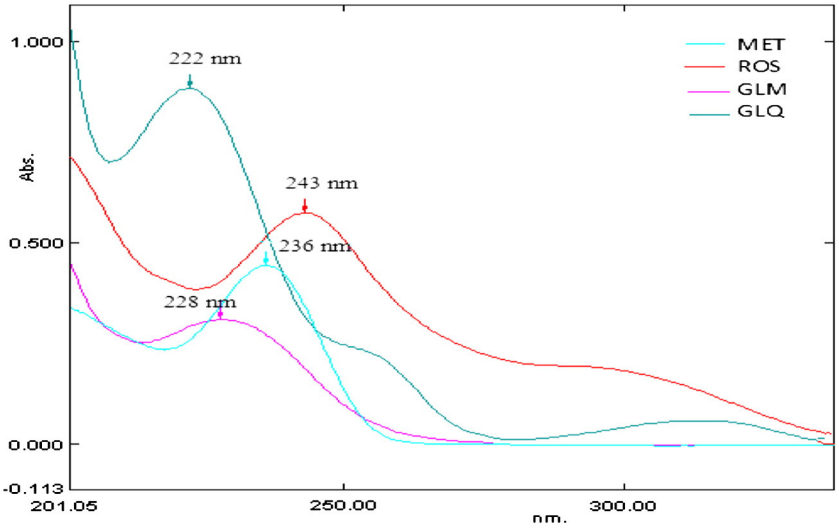

Fig. 2. Representative UV spectra of MET, ROS, GLM and GLQ showing their λ_{max}.