

## QUANTITATIVE ESTIMATION OF METFORMIN IN DIABETIC PATIENTS SERUM BY RP-HPLC

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### ABSTRACT

Metformin is an oral medication that lowers blood glucose (sugar) and is used for treating type 2 diabetes. Now a days maximum people using metformin according their prescription. After taking the drug some amount of that only released in to the body and remains in the blood. This study was done by quantitative estimation of remaining drug in the blood by RP-HPLC. According to the serum analysis the resultant drug in the blood was high amount in aged people when compare to younger people because of their metabolic activities.

**Keywords:** Metformin, metabolic, serum, RP-HPLC, diabetes.

### INTRODUCTION

The main use for metformin is in the treatment of diabetes mellitus type 2, especially in overweight people. In this group, over 10 years of treatment<sup>1-4</sup>, metformin reduced diabetes complications and overall mortality by about 30% when compared with insulin and sulfonylureas (glibenclamide and chlorpropamide) and by about 40% when compared with the group only given dietary advice. This difference held in the patients who were followed for 5–10 years after the study.

Since intensive glucose control with metformin appears to decrease the risk of diabetes-related endpoints in overweight diabetic patients, and is associated with less weight gain and fewer hypoglycaemic attacks than are insulin and sulphonylureas, it may be the first-line pharmacological therapy of choice in these patients. In addition, metformin had no effect on body weight: Over the 10-year treatment period, the metformin group gained about 1 kg, the same as the dietary advice group, while the sulfonylureas group gained 3 kg, and the insulin group, 6 kg. As metformin affords a similar level

of blood sugar control to insulin and sulfonylureas, it appears to decrease mortality primarily through decreasing heart attacks, strokes and other cardiovascular complications.

Metformin has a lower risk of hypoglycemia than the sulfonylureas, although it has uncommonly occurred during intense exercise, calorie deficit, or when used with other agents that lower blood glucose. Metformin is also not associated with weight gain, and modestly reduces LDL and triglyceride levels.

Metformin formula is  $C_4H_{11}N_5$  IUPAC Name is *N,N*-dimethylimidodicarbonimidic diamide. Molecular weight is 129.164 g/mol.

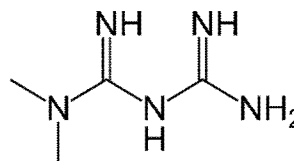


Fig.1: Chemical Structure of Carbohydrate

## EXPERIMENTAL

### Chemicals and Reagents

HPLC grade methanol and water were purchased from Merck Specialties Pvt. Ltd.

### Instrumentation and analytical conditions

The analysis of drug was carried out on a PEAK HPLC system equipped with a reverse phase C<sub>18</sub> column (250x4.6mm, 5µm in particle size), a LC-P7000 isocratic pump, a 20µl injection loop and a LC-UV7000 absorbance detector and running on PEAK Chromatographic Software version 1.06. Isocratic elution with methanol: water 70:30 (V/V) PH- 4.6 adjusted with ortho phosphoric acid was used at a flow rate of 1.4ml/min UV detection at 232nm. The mobile phase was prepared freshly and degassed by sonicating for 5 min before use.

### Collection of samples

From a local general hospital blood was collected from five members of diabetic patients those people are used metformin according to their prescription.

### Preparation of samples

From the blood serum was separated. 0.5ml of this serum was taken in a test tube and added 0.1ml of 1M NaOH and 5ml of dichloromethane and mixed about 20min in vortex mixer and centrifuged at 3000 rpm for 10min. From this centrifuged solution 4ml of organic layer was separated and evaporated to dryness to get residue. To this residue 100µl of 1M acetic acid and 3ml of n-Hexane and mixed for 5 min by vortex mixer and evaporated the organic layer and finally the remaining sample was injected into HPLC and chromatogram was recorded.

### Preparation of standard metformin

10mg of metformin was accurately weighed and it transferred into a 10ml volumetric flask and dissolved in mobile phase and make up the volume up to the mark with mobile phase (stock solution) and it sonicated for 5min. Finally 5µg/ml sample was prepared from stock solution. The sample was injected into HPLC and chromatogram was recorded.

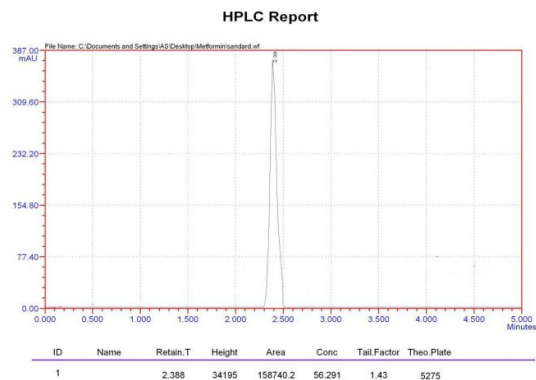


Fig.2: A Typical Chromatogram of Metformin

## RESULT AND DISCUSSION

From the experiment in different age group people the amount of metformin rest in the blood was different. Generally when the drug enter in to the body some amount of that consumed by the body the rest of drug is remaining in the blood.

Table 1

S. No.	Gender & Age	Amount of metformin found
Patient.1	Female 56yrs	0.67µg/ml
Patient.2	Male 45yrs	0.20µg/ml
Patient.3	Male 64yrs	0.748µg/ml
Patient.4	Male 27 yrs	0.093µg/ml
Patient.5	Female 48yrs	0.246µg/ml

## CONCLUSION

The drug is remains high amount in blood along with the age increment after absorption into the tissues.

## REFERENCES

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