

Non Incretin Effects of DPP-4 Inhibitors: Comparative Study

A.S.A.Bhatti¹, Aliya Shabbir², Abdul Waheed Shehzad³, M.A.Bhatti⁴

Department of Pharmacology, Shalamar Medical and Dental College Lahore^{1,2}. The Children Hospital and Institute of Child Health, Lahore³.

Department of Pharmacology, Avicenna Medical College, Lahore⁴.

ABSTRACT

Background: Diabetes mellitus is a metabolic syndrome that adversely affects all parts of the body especially cardiovascular system. Dipeptidyl peptidase-4 (DPP-4) is a multifunctional protein and its inhibition has diverse effects. DPP-4 inhibition was shown to improve survival rate after myocardial infarction in mice. Beneficial myocardial metabolic effects of DPP-4 inhibitors have been observed in experimental models. Cardiovascular outcome trials of DPP-4 inhibitors show variable adverse cardiovascular events.

Objective: This experimental study was aimed to study the direct cardiovascular effects of DPP-4 Inhibitors on chronotropicity (Heart Rate,HR), inotropicity (Apical Force and; dP/dt(max), ECG and Coronary Flow(C.F) and detect its potential useful and harmful effects on cardiovascular parameters.

Methods: The effects of graded doses (10^{-9} - 10^{-6} M) of Sitagliptin (S) and Vildagliptin(V) were observed on retrograde perfused isolated rabbit hearts with warm Krebs-Henseliet solution on Radnoti working heart system. Fifty four(54) rabbits were grouped into nine groups i.e ; I(S1), II(S2), III(S3), IV(S4), V(S5), VI(S6), VII(V1), and VIII(V2) and IX(V3) each comprising of six animals(n=6).Effects of experimental drugs were observed on chronotropicity(HR), inotropicity (Apical Force and Peak rate of rise of LVP i.e; dP/dt(max) and Coronary flow(CF). The results were statistically analyzed with Graph Pad Prism and wherever necessary paired or unpaired "t" test was applied.

Conclusion: Sitagliptin and Vildagliptin both have suppressant effects on HR. Sitagliptin has positive and Vildagliptin had negative inotropic effects. Both drugs (10^{-8} - 10^{-6} M); decrease coronary flow but have no significant effect on ECG.

Key Words: T2DM, IHD, DPP-4 Inhibitors, Apical force, dP/dt(max).

INTRODUCTION

Diabetes mellitus can result into Coronary Heart Disease (CHD) later on leading to an acute myocardial infarction^{1,2,3}. Gliptins have cardiovascular depressant effects as a result of inhibitory action on the dipeptidyl peptidase-4 enzyme^{4,5}. It has been observed that Sitagliptin reduces elevated cardiac fatty acid uptake and oxidation, prevents myocardial fibrosis and reduces oxidative stress in cardiomyocytes in small animals⁶. The role of DPP-4 inhibitors on the heart is still unclear⁷. In addition to the insulinotropic effect, GLP-1 signaling has been reported to modulate cardiac functions. In animals DPP-4 inhibition has shown to improve survival rate after myocardial infarction but the precise mechanism is not known. There is also need of comparative study of direct cardiovascular effects of DPP-4 Inhibitors.

MATERIALS AND METHODS

This experimental study was carried out in the laboratory of the Pharmacology Department, Shalamar Medical and Dental College Lahore after the approval of the project from Institutional Review and Ethical Board (IRB) of Shalamar Medical and Dental College, Lahore and according to the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health (National Institutes Of Health Publication 85-23, revised 1996).

Grouping of Animals: Fifty four (54) rabbits were grouped into seven groups i.e; I(S1), II(S2), III(S3), IV (S4), V (S5), VI (S6), VII (V1), VIII (V2) and IX(V3) each comprising of six animals (n=6)

Note: Groups 1, 11, 111, V1, V, V1 were treated with four concentrations or doses i. 10^{-9} M, 10^{-8} M, 10^{-7} M, 10^{-6} M while Groups V11, V111 and 1X were treated with 10^{-8} M, 10^{-7} M, and 10^{-6} M.

Preparation of Heart for Perfusion: 54 normal male rabbits (weighing between 1.5-2 kg) were purchased from the local market. Each rabbit was injected 5000IU (1ML) of heparin through the marginal ear vein one hour before slaughtering. Heart was removed from the chest after midsternal incision and immediately placed in an ice cold Krebs-Henseleit solution. The heart was mounted on Langendorff working heart system via aortawhose stump was secured about 3 mm on the grooved perfusion cannula by a silk ligature. A weight was attached to the heart apex and left to hang freely exerting resting tension of one gram. Heart was perfused in retrograde at a constant flow of 52ml/min with warm Krebs-Henseleit solution. It was allowed to stabilize for 30 mins in the perfusate which was equilibrated with 95%O₂ and 5% CO₂ with Krebs-Henseleit solution with pH of 7.35 at 37°C. A saline-filled balloon catheter was inserted into the left ventricle which was often used to measure isovolumetric work. Balloons slightly larger than the maximum expanded volume of the ventricle were used to avoid effects of measuring the resistance of the balloon to stretch.

Drugs Used: Oral hypoglycemic was procured from the Wilshire pharmaceutical industry in the powder form free from any base. Following drugs were employed: Sitagliptin(S) and Vildagliptin(V)

RESULTS

1. Sitagliptin (10⁻⁹ - 10⁻⁷M) and Vildagliptin (10⁻⁸ - 10⁻⁴M) have suppressant effects on HR (Fig.1).
2. Sitagliptin (10⁻⁹ - 10⁻⁴M) has positive and Vildagliptin (10⁻⁸ - 10⁻⁴M) negative inotropic effects (Fig.2, 3).
3. Both drugs (10⁻⁸ - 10⁻⁶M) decrease coronary flow (Fig.4). However, Sitagliptin (10⁻⁴M) is increases coronary flow.
4. There was no significant effect on ECG (R and ST-segments) of individual drugs (Table 1).
5. There was significant difference in R and ST-segments when the results of two drugs were compared.

It means with decreased heart rate, coronary flow was also decreased with administration of both drugs. It seems that both drugs have direct

suppressants effects on conducting tissue and myocardium and cause decreased coronary flow. This effect is also demonstrated by Vildagliptin in the form of significant decreased contractility of myocardium. Sitagliptin has positive inotropicity (Table 1).The results were statistically analyzed with graph Pad prism and wherever necessary paired or unpaired “t” test was applied.

Effects of Drugs on Heart Rate, Apical Force, Max dp/dt, Coronary Blood Flow

Fig. 1

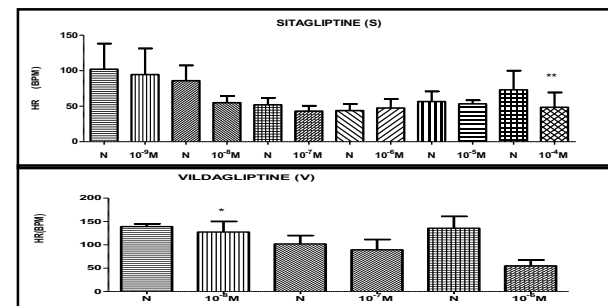


Fig: 2

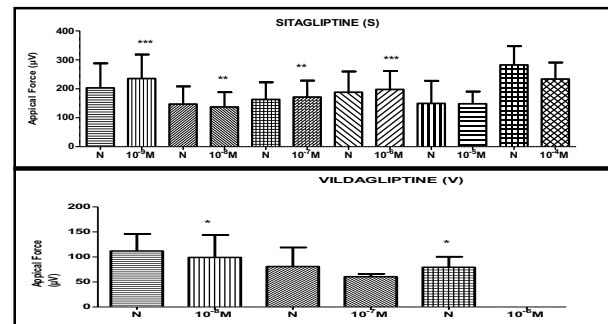


Fig: 3

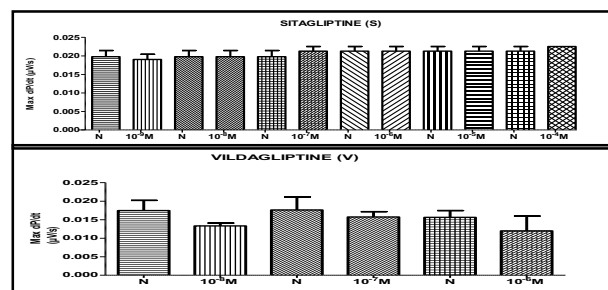


Fig: 4

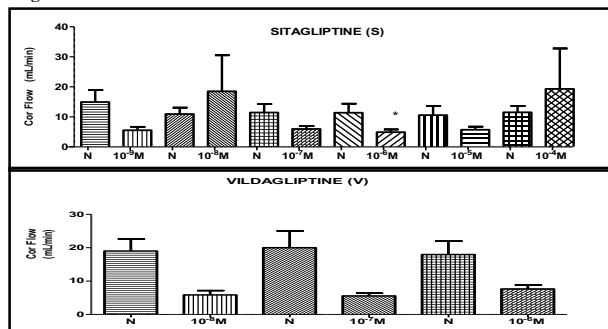


Table No: 1

Comparative Study of Effects of Sitagliptin (S) and Vildagliptin (V)			
Drugs		Mean ± SE	"p" Value
Heart Rate (BPM)			
SITAGLIPTIN	S (10 ⁻⁸ M)	111.5 ± 6.611	p>0.05
VILDA GLIPTIN	V (10 ⁻⁸ M)	113.8 ± 22.84	
SITAGLIPTIN	S (10 ⁻⁷ M)	111.2 ± 6.680	p>0.05
VILDA GLIPTIN	V (10 ⁻⁷ M)	92.90 ± 24.77 N=5	
Apical Force (μV)			
SITAGLIPTIN	S (10 ⁻⁸ M)	137.3 ± 50.94	p>0.05
VILDA GLIPTIN	V (10 ⁻⁸ M)	99.00 ± 44.58	
SITAGLIPTIN	S (10 ⁻⁷ M)	171.6 ± 56.88	p>0.05
VILDA GLIPTIN	V (10 ⁻⁷ M)	60.60 ± 5.793	
Max dP/dt (μV/s)			
SITAGLIPTIN	S (10 ⁻⁸ M)	0.019 ± 0.0016	p<0.05**
VILDA GLIPTIN	V (10 ⁻⁸ M)	0.012 ± 0.0006	
“R” Wave Amplitude (mV)			
SITAGLIPTIN	S (10 ⁻⁸ M)	1.285 ± 0.387	p>0.05
VILDA GLIPTIN	V (10 ⁻⁸ M)	226.4 ± 161.2	
SITAGLIPTIN	S (10 ⁻⁷ M)	1.294 ± 0.387	p<0.05***
VILDA GLIPTIN	V (10 ⁻⁷ M)	588.1 ± 108.4	
“ST” Height (mV)			
SITAGLIPTIN	S (10 ⁻⁸ M)	0.3105 ± 0.118	p<0.05*
VILDA GLIPTIN	V (10 ⁻⁸ M)	434.8 ± 177.1	
SITAGLIPTIN	S (10 ⁻⁷ M)	0.3118 ± 0.118	p>0.05
VILDA GLIPTIN	V (10 ⁻⁷ M)	75.70 ± 162.8	
“QT” Interval (ms)			
SITAGLIPTIN	S (10 ⁻⁸ M)	19.34 ± 19.21	p>0.05
VILDA GLIPTIN	V (10 ⁻⁸ M)	21.10 ± 0.690	

p* value< 0.05 was considered statistically significant

DISCUSSION

Cell surface antigen CD26 is synonym of Dipeptidyl peptidase-4 (DPP-4). Its role in metabolic disorders, neoplasia and inflammatory conditions is well known. It is due to its proteolytic activity. DPP-4 is also present in kidney, small intestine, liver and heart tissues^{8,9}. DPP-4 break down products become soluble in body fluids¹⁰. Endogenous levels of glucagon-like peptide-1 (GLP-1) levels are raised by DPP-4 Inhibitors. GLP-1 is responsible for lowering blood glucose

level. It was hypothesized that treatment with DPP-4 inhibitors is cardio protective¹¹. There are certain endogenous substances like stromal derived factor-1 (SDF-1), a known chemokine substrate of DPP-4 and other gastrointestinal hormones and cytokines which have shown cardio protective effects in experimental studies¹². Sitagliptin reduces plasma glucose rise, body weight, myocardial fibrosis and oxidative stress¹³. In our experiment increased chronotropicity and decreased coronary flow may

be due to release of these cytokines. Opposite effect of Sitagliptin and Vildagliptin on inotropy may be due to changes in the intracellular calcium on account of their different chemical structure and their influence. Sitagliptin antagonized the antihypertensive effect of enalapril in one of the experiments. This was considered to be due to activation of the sympathetic nervous system by substance P and neuropeptide Y which is responsible for vasodilatory effects¹⁴. Dipeptidyl peptidase (DPP-4) inhibitors can exert adverse cardiovascular effects. DPP-4 inhibitors may stimulate cAMP in cardiomyocytes¹⁵. Several experimental studies, clinical trials and meta-analysis have shown no conclusive results about risks and benefits of DPP-4 Inhibitors^{16,17}.

CONCLUSION

DPP-4 Inhibitors may cause cardiovascular effects due to their non-incretin properties when prescribed. Moreover, their interaction with other antihypertensive like ACE inhibitors must be kept in view when prescribing sitagliptin which in our study has shown positive inotropic effects.

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Corresponding Author:

A Shabbir Ali Bhatti
 Department of Pharmacology,
 Shalamar Medical and Dental College, Lahore.
Email: shabbiralibhatti@gmail.com