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EFFECT OF ALLICIN ON PHARMACOKINETICS AND PHARMACODYNAMICS OF REPAGLINIDE IN NORMAL AND STREPTOZOCIN INDUCED DIABETIC RATS

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ABSTRACT

Along with prescribed drugs, diabetic patients are using different phytochemicals/herbs having antidiabetic properties. In these cases there is chance of alterations in pharmacokinetics and pharmacodynamics of drug due to interactions. Repaglinide is a meglitinide class of oral antidiabetic drug and it was metabolized by CYP2C8, CYP2C9 and CYP3A4 enzymes. The present study investigates that interaction of repaglinide with allicin. The rats were divided into normal and diabetic groups, studied for different pharmacokinetic parameters like C_{max}, AUC_{0-n}, AUC_{total}, $t_{½}$, MRT and the clearance, V_d and pharmacodynamics like body weight, blood glucose level. The result indicates that combination of repaglinide with allicin, improves Cmax, AUC and MRT where as Volume of distribution and clearance decreases. Repaglinide metabolism was decreased with inhibitory action of allicin on CYP3A4. Finally repaglinide in combination with allicin, shows more control on blood glucose levels of diabetic rats.

Keywords: Allicin, CYP enzymes, Repaglinide, Pharmacokinetics and pharmacodynamics

INTRODUCTION

Diabetes mellitus (**DM**) is the most common metabolic disorder and it leads to increased sugar levels in blood and this is mainly due to insufficient production of insulin from pancreas **[1]**. There are many oral antidiabetic drugs which are used by diabetic patients; along with these some herbal drugs/phytochemicals were also used.

Combination of drugs and herbs may cause interactions and those are showing neither beneficiary effect nor adverse effect on the patient **[2]**. For this reason it is important to study the effect of herb-drug interactions and knowing the information of how they alter the pharmacokinetics and pharmacodynamics parameters of drug **[3]**.

Repaglinide is an oral antidiabetic agent belongs to meglitinide class and it lowers the blood glucose level by stimulating the release of insulin from pancreas. Repaglinide was mainly metabolized by CYP2C8, CYP2C9 and CYP3A4 enzymes and after metabolism drug was converted into inactive metabolites **[4]**.

Allicin is obtained from Allium sativum (Amaryllidaceae), commonly known as garlic, it possess antidiabetic properties and largely used in Indian foods. Some in-vitro reports showed that allicin has inhibitiory action on CYP enzymes particularly CYP3A4, CYP2C8, CYP2C9 [**5 -6]**. So, it is necessary to study the effect of allicin with repaglinide and knowing the changes in different pk and pd parameters.

MATERIALS AND METHODS

Repaglinide was obtained as gift sample from Novartis, Hyderabad and Allicin was purchased from yucca enterprises, Mumbai. Streptozocin was supplied from sisco labaratories, Methanol HPLC grade supplied from Merck chemicals, Mumbai. All the chemicals used for the study are of analytical grade.

Maintenance of animals:

After obtaining the approval from institutional animal ethical committee (CPCSEA Reg.no. IAEC/ 14/ UCPSc/ KU/2020), male albino rats of wistar strain were purchased from Sainath Agencies, Hyderabad, weighing 200-250gm used for the study. The animals were housed under standard laboratory conditions (equal proportion of day and night cycle in a day, at ambient temperature and fed with standard rat pellet diet **[7]**.

HPLC analysis of Repaglinide:

Repaglinide was estimated by previously reported HPLC method **[8]**. The system consisted of binary LC-20AD pumps with a micro gradient mixer. RP C18 column, 250 mm×4.6 mm, 5 µm (Phenomenex Luna) was used at 23±2.0 °C. All the operations and analysis of data obtained were controlled by lab solutions software. A mixture of methanol and distilled water in the ratio of 80:20 (v/v) used as mobile phase and delivered at a flow rate of 1 ml/min. The mobile phase was degassed by sonicator and filtered through 0.22 µm membrane filter. Then detection was carried out at a

wavelength of 260 nm. The total run time of the method was set at 10 minutes.

Experimental design

Grouping of normal rats and PK study

Overnight fasting, rats were divided into two groups (n = 6). Group I was administered with repaglinide (0.2mg/kg) suspended in normal saline on the 8th day. Group II was pretreated with allicin (8 mg/kg) for 7 days and on the 8th day with repaglinide (0.2 mg/kg) followed by allicin **[9]**. Through retro orbital plexus blood samples were collected by using heparinised capillary tubes, and immediately same volume of normal saline was replaced intra peritoneally. The blood was collected at time intervals of 0.5, 1, 2, 4, 8, and 24 hrs in every group. Serum was separated after centrifugation at 8000 rpm for 15 min and the samples were stored at –20°C until analysis. The pharmacokinetic parameters like C_{max}, T_{max}, AUC_{0-n}, AUC_{total}, $t_{1/2}$, MRT, Cl and V_d were calculated.

Induction of diabetes in rats

Diabetes was induced by using STZ (55 mg/kg) in citrate buffer (pH 4.5) to the overnight fasted Wistar rats. After 72 h, blood samples were collected from rats by retro-orbital puncture and the serum was analyzed for glucose levels. Animals with blood glucose level >250 mg/dL were considered as diabetic and were used for the study **[10]**.

Grouping of diabetic rats and PK study:

Diabetic rats were divided into two groups (n = 6) and were treated same as normal rat group, blood samples were collected at different time intervals. The samples were stored at appropriate storage conditions and samples were analyzed by using HPLC.

Pharmacodynamic study

The STZ induced diabetic rats were fasted overnight and divided into 4 groups (n = 6). The animals of group I (diabetic control, normal saline), group II (Repaglinide, 0.2mg/kg), group III (Allicin, 8mg/kg) and group IV [Repaglinide, 0.2mg/kg+ Allicin, 8mg/kg] were treated orally. The effect of the repaglinide, allicin alone and their combinations on blood glucose level was studied up to 24 h. Blood samples were drawn from the retro-orbital plexus of the rats at 0 (Initial fasting blood sample),0.5, 2, 4, 8, 12 and 24h after the treatment. The samples were analyzed for blood glucose using glucose oxidase-peroxidase method and percentage reduction in blood glucose concentrations was determined **[11]**.

% glucose reduction at t hour = [(A-B) / A] × 100 A = mean glucose levels at t hour B = mean glucose levels at 0 hour.

Statistical analysis: The Pharmacokinetic parameters were calculated by using Kinetica TM software (version 4.4.1). All values of pharmacokinetic and pharmacodynamic studies were expressed as Mean±SD. The data were statistically evaluated using one-way analysis of variance (ANOVA). Results were considered to be statistically significant when $p \leq 0.05$.

RESULTS & DISCUSSION

Pharmacokinetics of repaglinide in normal and diabetic rats:

In normal and diabetic pretreated rats, compared with the control group (given repaglinide alone), the co-administration of allicin significantly ($p<0.01$) increased Cmax (1.43 and 1.91 times), AUC_{0-n}(1.56 and 2.53 times), AUC_{total} (1.97 and 3.05 times), $t_{1/2}$(1.52 and 1.73 times), MRT (0.74 and 1.23 times), whereas, the clearance (1.14 and 2.36 times) and volume of distribution (1.52 and 2.09 times) of repaglinide was decreased. The Tmax was not altered significantly in both normal and diabetic rats. Combination of herb-drug, leads to increase in pharmacokinetic parameters like C_{max}, AUC_{0-n}, AUC_{total}, $t_{1/2}$ and MRT, decrease in volume of distribution and clearance due to the changes in metabolism of repaglinide by inhibitory action of allicin on CYP enzymes **[12, 13]**.

Pharmacodynamic study in different groups

The body weight of diabetic control (group I) was gradually decreased, whereas in all other groups it was increased gradually from 7 days onwards to 28 days, which showed that these herb and drug do not have much effect on degradation of depot fat to maintain the body weight.

The mean serum glucose level and percentage glucose reduction study of diabetic rats and the data indicates that there is a maximum reduction of serum glucose level in combination of repaglinide with allicin treated groups (48.9%), when compared with standard (repaglinide, 38.36%), allicin (33.6%), alone pretreated groups at the 4^{th}h, respectively. However, combination of allicin with repaglinide showed significant decrease ($p\leq0.001$) in serum glucose level at different time intervals **[14]**.

Table 1: Mean pharmacokinetic parameters of repaglinide in different groups of normal and STZ-induced diabetic rats.

PK parameter	Repaglinide		Repaglinide+Allicin	
	Normal	Diabetic	Normal	Diabetic
C_{max} (µg/ml)	0.852±0.0531	2.26±0.024	2.4±0.162**	4.6±0.183**
T_{max} (h)	1.14±0.346	1.1	1	1
AUC_{0-n} (h.µg/ml)	25.26±1.64	34±1.83	32±2.31*	85±3.57**
AUC_{total} (h.µg/ml)	27±2.64	53±3.82	37±2.71*	113±4.83**
$t_{½}$ (h)	1±0.26	1.5±0.31	2±0.28	3.5±0.39
MRT (h)	3±0.18	4±0.32	4.5±0.27	5.5±0.41
CL (ml/h/kg)	16.8±1.04	9.3±1.75	11.35±1.28**	5.63±2.05**
V_d (ml/kg)	41±2.05	30±1.53	25±2.38**	12±1.74**

All values are expressed as mean ± SD (n=6)
***p<0.05, **p<0.01 considered as significant when compared with control groups**

Table 2: Effect of Repaglinide, Allicin and combination of both on body weights in STZ - induced diabetic rats (28 days)

	Body weight in grams				
Group	1st day	7th day	14th day	21st day	28th day
I (Control)	230.1±3.12	206.7±4.2	184.3±1.5	158.5±2.5	137.8±5.2
II(RG)	210.4±2.5	235.3±5.4	251.5±3.8	268±2.9	281.5±3.4
III(Allicin)	205±4.23	230.4±3.2	260±2.4	274.3±5.2	295.3±1.8
IV(RG+Allicin)	206.7±3.64	228.9±2.5	254.3±3.7	284±3.8	303.4±3.6

Table 3: Comparison of mean serum glucose levels and percentage reduction of serum glucose level of different groups of STZ-induced diabetic rats

Group	Treatment	Dose (mg/kg)	Blood glucose level (mg/dl) at different time intervals							
			0 h	0.5 h	1 h	2 h	4 h	8 h	12 h	24 h
I	Control	--	320±4.26	318±3.71 (0.625%)	316±2.37 (1.25%)	317±1.87 (0.93%)	319±3.72 (0.312%)	318±2.65 (0.625%)	320±3.06 (0.13%)	319±2.74 (0.312%)
II	RG	0.2	318±3.68	290±2.53 (8.8%)**	262±2.81 (17.6%)**	227±1.85 (28.6%)**	196±2.37 (38.36%)**	235±1.36 (26.1%)**	270±2.85 (15.9%)**	305±2.15 (4.08%)**
III	Allicin	8	324±3.15	296±2.74 (8.64%)**	268±1.72 (17.28%)**	240±3.62 (25.9%)**	215±2.68 (33.6%)**	258±1.83 (20.3%)**	290±2.46 (10.4%)**	310±1.73 (4.32%)**
IV	RG + Allicin	0.2+8	321±2.85	285±1.83 (11.21%)**	236±3.62 (26.4%)**	194±2.85 (39.56%)**	164±3.83 (48.9%)**	190±3.85 (40.8%)**	238±1.62 (25.8%)**	283±3.64 (11.83%)**

RG: Repaglinide. All values are expressed as mean ± SD (n=6)
***p<0.05, **p<0.01 considered as significant when compared with control groups**

CONCLUSION

Combination of repaglinide and allicin showed good hypoglycemic action in diabetic rats. This may be due to inhibition of CYP3A4 enzyme by allicin and there by repaglinide concentration was increases. Hence this particular herb-drug combination has beneficiary effect and need special attention while using by diabetic patients.

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REFERENCES

[1] Archana M.P and Gurupadayya BM (2018). Potential Herb Drug Interactions in antidiabetic Drugs and Herbal Drugs. International Journal of Phytopharmacy; 8 (6): 65-72.

[2] Tripathi P, Gupta PP and Lal VK (2013) Interaction of Momordica charantia with metformin in diabetic rats. American Journal of Pharmacology Toxicology; 8: 102-106.

[3] Saurabh Nimesh, Ravi Tomar and Shubham Dhiman (2019). Medicinal Herbal Plants and Allopathic Drugs to Treat Diabetes Mellitus: A glance; Advances in Pharmacology and Clinical Trials. 4(1).

[4] Jyothi P, Thirupathi G and Narsimhareddy Y (2017). Pharmacokinetic and pharmaco-dynamic interaction study of curcumin with repaglinide in normal and diabetic rats. Journal of global trends in pharmaceutical sciences; 8(3): 4130 – 4137.

[5] Thomson M, Al-Qattan KK, Js D and Ali M (2016). Anti-diabetic and antioxidant potential of aged garlic extract (AGE) in streptozotocin induced diabetic rats. BMC Complement Alternative Medicine; 16:17.

[6] Foster BC, Foster MS, Vandenhoek S, Krantis A, Budzinski JW, Arnason JT, Gallicano KD and Choudri S (2001). An in-vitro evaluation of human cytochrome P450 3A4 and P-glycoprotein inhibition by garlic. Journal of Pharmaceutical sciences; 4:176–184.

[7] Liu Y, Qi H, Wang Y, Wu M and Cao Y (2012). Allicin protects against myocardial apoptosis and fibrosis in streptozotocin-induced diabetic rats. Phytomedicine, 19(8-9): 693-698.

[8] Seema MD, Pramod BK, Nikhil DA. (2014) Comparison of UV spectrophotometry and high performance liquid chromatography methods for the determination of repaglinide in tablets. Journal of Pharmaceutical analytical methods, 3: 68-72.

[9] Umachandar L, Habibuddin M and Sivakumar R (2018) Effect of Allicin on Pharmacodynamics and Pharmacokinetics of Gliclazide in Diabetic Animal Models. Journal of Complement Medicine Alternate Healthcare; 8(1).

[10] Sharma, A., B.L. Fish, J.E. Moulder, M. Medhora, J.E. Baker, M. Mader and E.P. Cohen (2014) Safety and blood sample volume and quality of a refined retro-orbital bleeding technique in rats using a lateral approach. Lab. Anim., 43: 63-66.

[11] Cheguri S, Ajmera RR and Ciddi V (2017) Pharmacokinetic and Pharmacodynamic Interaction of Quercetin with Saxagliptin in Normal and Diabetic Rats. Pharmacologia. 8 (3): 90-94.

[12] Akbarzadeh, A., D. Norouzian, M.R. Mehrabi, S. Jamshidi and A. Farhangi (2007) Induction of diabetes by streptozotocin in rats. Indian Journal Clinical Biochemistry; 22: 60-64.

[13] Trinder P (1969) Determination of blood glucose using 4- amino phenazone as oxygen acceptor. Journal of Clinical Pathology; 22: 246.

[14] Chandra SM and Jaya CRP (2012) Effect of simvastatin on the pharmacodynamic activity of repaglinide in rats/rabbits. International Journal of Diabetes Research; 1: 52-57.