

Influence of Itraconazole on Sulfonylureas-Induced Hypoglycemia in Diabetic Rats

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Antifungals like fluconazole, ketoconazole and miconazole have been reported to interact with sulfonylurea antidiabetic agents and potentiate their hypoglycemic effect. In the present investigation we have studied interaction of itraconazole, with tolbutamide and glibenclamide in alloxan-induced diabetic rats. In the first set, the *per se* effect of itraconazole (30 mg/kg, po) on blood glucose levels was studied. In the second set, the hypoglycemic activity of tolbutamide (40 mg/kg, po) and glibenclamide (40 µg/kg, po) was studied in separate groups of animals. After 1 w of washout period, these animals were treated with itraconazole (30 mg/kg, po) for 7 d. On 8th d, 1 h after itraconazole administration, the effect of sulfonylureas was studied as usual on blood glucose levels.

Patients with diabetes mellitus have an increased pre-disposition to dermatophytic infections and these infections disrupt the skin integrity, provide an avenue for bacterial super infections¹. Hence, diabetics with either dermatophytic or other fungal infections should be promptly treated with a suitable antifungal agent for a specified period. However, during such therapy there is every possibility of interaction between the antifungal agents used with the routine oral antidiabetic drugs². Further, drug interactions have been reported to be the fourth to sixth leading cause of death in hospitalized patients in United States³. This alarming report warrants the immediate necessity of study to understand the gravity of drug interactions among the probable multi-drug prescriptions.

Antifungal agents like ketoconazole⁴, fluconazole⁵ and miconazole⁶ have been reported to interact with

sulfonylureas and potentiate their hypoglycemic effect. Itraconazole an antifungal agent, active orally, with reduced toxicity, better pharmacokinetics and broader antifungal spectrum. However, it has been reported that itraconazole inhibits the metabolism of concomitantly used drugs like warfarin⁷, cyclosporine⁸ and terfenadine⁹. But, its interaction with sulfonylureas is not well established in diabetic animals. Hence, the present study was planned to understand the possible interaction between itraconazole and sulfonylureas (tolbutamide and glibenclamide) in alloxan-induced diabetic rats.

MATERIALS AND METHODS

The study was under taken with due approval of the study protocols by the Institutional Animals Ethical Committee. Wistar rats of either sex weighing between 150-260 g were housed in clean polypropylene cages in air conditioned animal house of the institution and fed with commercial pelleted rat chow diet and water was given *ad libitum*. Itraconazole (Glenmark Pharmaceuticals, Nasik), tolbutamide (Albert David, Mumbai), glibenclamide and glucose estimation kit of Dr. Reddy's Laboratories,

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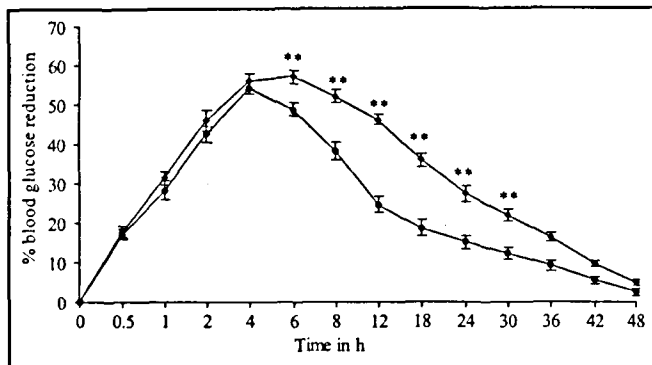


Fig. 1: influence of itraconazole on tolbutamide-induced hypoglycemia in diabetic rats

Each legend represents mean \pm SEM, n=6 and **represents significant at $p < 0.01$. Percentage blood glucose reduction with tolbutamide (-●-) and itraconazole + tolbutamide (-◆-)

Hyderabad were used in the study.

Induction of diabetes:

Rats were injected with alloxan (150 mg/kg) subcutaneously¹⁰. From d 2 onwards the blood samples were drawn and blood glucose levels were determined to confirm the development of diabetes. Rats exhibiting blood glucose levels of 240 mg/100 ml and above were considered as diabetic and selected for the study.

Drug treatment:

Itraconazole, tolbutamide and glibenclamide were suspended separately in 5% (w/v) gum acacia suspension in distilled water and fresh suspensions were administered orally. All the animals used in the study were fasted for 18 h prior to individual drug administration.

Effect of itraconazole:

In the first set of experiments the effect of itraconazole (30 mg/kg, po) was studied on blood glucose level in alloxan-diabetic rats (n=6). In the second set of experiments, alloxan-diabetic rats were divided into two groups; one treated with tolbutamide (40 μ g/kg, po) and second with glibenclamide (40 μ g/kg, po). These animals were fasted for 18 h prior to drug administration. They received tolbutamide or glibenclamide and blood samples were collected from the tail vein at the intervals of 0, 0.5, 1, 2, 4, 6, 8, 12, 18, 24, 30, 36, 42 and 48 h and blood glucose levels were determined¹¹. The animals were allowed one week wash out period to eliminate the administered drugs completely.

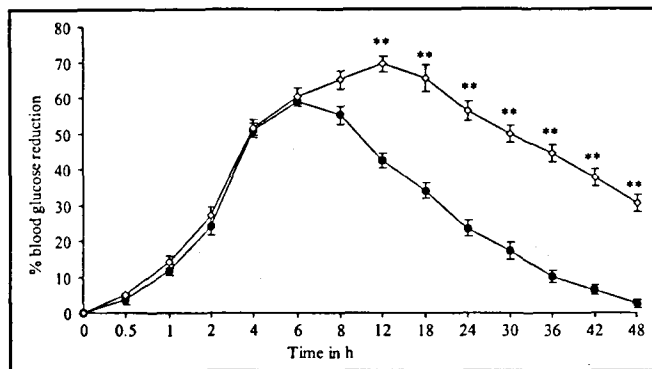


Fig. 2: influence of itraconazole on glibenclamide-induced hypoglycemia in diabetic rats

Each legend represents mean \pm SEM, n=6 and **represents significant at $p < 0.01$. Percentage blood glucose reduction with glibenclamide (-●-) and (-◇-) itraconazole+glibenclamide

After 1 w of washout period, the animals of both the groups were treated with itraconazole (30 mg/kg/d, po) for 7 days. During this period the animals had free access to food and water. On 7th d, 6 h after the itraconazole administration, food was deprived and the animals were fasted for 18 h and water was given *ad libitum*. On 8th d, 1 h after itraconazole administration, the animals of both the groups received the above sulfonylureas as usual. Blood samples were collected thereafter at above mentioned intervals and blood glucose levels were estimated¹¹. The hypoglycemic activity of tolbutamide and glibenclamide at any required time 't' was calculated as the percentage blood glucose reduction at that time with respect to initial blood glucose level.

Statistical analysis:

The values are presented as mean \pm SEM. The data was analyzed by using One-way Analysis of Variance (ANOVA) followed by Dunnett's multiple comparisons and considered statistically significant at $P < 0.01$.

RESULTS

Results of the study have been graphically depicted in figs. 1 and 2. For the assessment of hypoglycemia, the onset of action (time taken to produce 20% reduction in blood glucose level), the peak effect and the duration of hypoglycemia (duration during which at least of 20% reduction in blood glucose levels were maintained) were considered. Itraconazole (30 mg/kg, po) treatment alone did not alter the blood sugar levels.

Itraconazole pre-treatment did not produce much

alteration in tolbutamide-induced hypoglycemia. There was change in peak effect from $54.2 \pm 1.64\%$ to $57.0 \pm 1.66\%$, with prolongation of duration of hypoglycemic action from 12h to about 30h.

On the other hand, itraconazole pretreatment produced very significant alteration in glibenclamide-induced hypoglycemia. The peak effect was significantly increased from $59.0 \pm 1.25\%$ at 6th h to $69.3 \pm 2.31\%$ at 12th h ($p < 0.01$) and there was prolongation of hypoglycemic action from around 24h to more than 48h. Out of the six animals studied, one animal died of severe hypoglycemia at 24th h and others suffered hypoglycemic convulsions and recovered. However, there was no change in the onset of action of either of the sulfonylureas.

DISCUSSION

Drug interactions involving induction and inhibition of microsomal enzymes are of scientific interest and clinical importance. Several studies have suggested the important role of hepatic cytochrome P-450 enzyme system in various pharmacokinetic type of drug interactions¹². Such interactions result in alteration of pharmacokinetic parameters and therapeutic efficacy of concomitantly used drugs and hence, require adjustment of their dosage schedule accordingly.

Sulfonylureas are metabolized by cytochrome P-450 2C9 enzyme system¹³⁻¹⁵, and some of the agents used to either treat or manage the co-existing diseases in Type 2 diabetes have been reported to interact with sulfonylureas by either inducing or inhibiting the enzyme responsible for their metabolism, and alter their action. Drugs like rifampicin¹⁶, phenytoin¹⁷ and phenobarbitone¹⁸ have been reported to induce cytochrome P-450 enzyme system and reduce the efficacy of sulfonylureas. On the other hand, cimetidine¹⁹, ranitidine²⁰, lansoprazole²¹, metronidazole²², ketoconazole, fluconazole and miconazole have been reported to inhibit the cytochrome P-450 enzyme system and retard the metabolism of sulfonylureas leading to hypoglycemia. In the present study, itraconazole pretreatment was found to significantly enhance the peak effect and duration of action of tolbutamide and glibenclamide induced hypoglycemia. Particularly with glibenclamide, there was severe drug interaction, wherein one animal died because of severe hypoglycemia, others exhibited hypoglycemic convulsions and recovered.

The documented reports indicate that, the drugs like phenytoin, warfarin and tolbutamide are metabolized by

the same isoenzyme and the inhibitors of this isoenzyme retard the metabolism of all these drugs and there by exhibit a similar spectrum of interactions^{23,24}. Itraconazole has been reported to inhibit the metabolism of warfarin, cyclosporine and terfenadine. Hence, according to the above reports, as itraconazole inhibits the metabolism of warfarin, it is also likely to inhibit the metabolism of sulfonylureas. Based on the above discussion, it may be concluded that, the potentiation of antidiabetic activity of sulfonylureas by itraconazole could be attributed to its inhibitory effect on cytochrome P-450 enzyme system responsible for their metabolism.

Itraconazole though inhibits the metabolism of both tolbutamide and glibenclamide, the incidence of hypoglycemia was very severe with glibenclamide. The probable mechanism and explanation for this exaggerated effect despite enzyme inhibition demands the metabolites estimation studies.

It is evident from the present study that, co-administration of itraconazole and sulfonylurea (particularly glibenclamide) may give rise to clinical emergency leading to severe hypoglycemia. This is particularly important in elderly subjects, since, hypoglycemia can present as an acute neurological emergency that may mimic cerebrovascular accident. So, the dose and/frequency of sulfonylurea administration should be readjusted accordingly, when they need to be co-administered with itraconazole to avoid fatal hypoglycemia.

The present investigation suggest the need of further studies to confirm and establish the influence of itraconazole on the pharmacokinetic parameters of concomitantly administered sulfonylureas, particularly glibenclamide in healthy and diabetic human subjects.

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