CARDIOVASCULAR SIDE EFFECT REMOTELY RELATED TO NSAIDs: A COMPARATIVE EXPERIMENTAL STUDY ON ALBINO RATS

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ABSTRACT

Non-steroidal anti-inflammatory drugs (NSAIDs) inhibit enzyme cyclo-oxygenase (COX)-1&2, required for anti-inflammatory effect, which comes along with side effect of gastric ulceration related to inhibition of COX-1. Whereas recent NSAIDs act selectively on COX-2 isoform. Thus referred as selective NSAIDs. Therefore selective NSAIDs are more beneficial than earlier non-selective NSAIDs in regard of incidence of gastric ulceration. Metabolism of these drugs is associated with liver and kidney. Therefore, we observed the comparative adverse effects of diclofenac sodium (diclo) a non-selective NSAIDs and valdecoxib (valde), a selective NSAIDs on renal function. The experiment was carried on 50 albino rats of duckrey (DR) strain, in equal sex ratio. Animals were divided into five groups (n=10). Group A, B1, B2, C1 & C2. Group A served as control, B1 & B2 were given diclo 5 & 10 mg/kg/day per orally, C1 & C2 were given valde 1& 2 mg/kg/day per orally respectively, for 30 days. Renal histological examination and biochemical parameters were estimated. We estimated serum creatine, blood urea nitrogen (BUN), uric acid (UA), Na+ & K+ for renal function. Kidney sections of all the groups showed thickening of glomerular basement membrane, increase in glomerular cellularity, and degeneration of proximal and distal convoluted tubule. Tubule in medulla showed degeneration with intraluminal protein exudates. Histopathological changes were more significant with valdecoxib. In biochemical estimation lower dose of diclo showed increase in serum K+ only, on other hand lower dose of valde showed more significant change and there was increase in serum creatine, BUN & K+. Whereas higher doses of diclo and valde showed highly significant increase in serum creatine, BUN & K+. Increased UA was observed only with higher dose of valde. Thus these result show renal insufficiency, fluid overload and accumulation of urea & K+. These can lead to congestive heart failure, pericarditis, hypertension, arrhythmias. Thus, chronic renal failure patient suffers from accelerated atherosclerosis and high incidence of cardiovascular disease can be explained. Valdecoxib though banned due to cardiovascular effects and atherosclerosis which can also linked indirectly from renal insufficiency.

Key words: NSAIDs, renal insufficiency, cardiac insufficiency.

INTRODUCTION

COX-2 selective inhibitors have been introduced since 1999 as an alternative to traditional non-steroidal anti-inflammatory drugs (NSAIDs). The COX enzyme exists in two isoforms COX-1& 2 (Needleman & Isakson 1997)[1], COX-1 mainly associated with homeostasis and its antagonism would result into gastric ulceration. Whereas inducible isoform is COX-2 and its antagonism thus provides relieve from pain and inflammation (Masferrer et al.1994[2], Seibert et al. 1994[3] and Warner et al. 1999)[4].

Recent COX-2 selective agents have been increased by the addition of valdecoxib, etoricoxib and lumiracoxib. These COX-2 selective drugs together with the NSAIDs cover a wide range of selectivities toward COX-1 and COX-2 (Van Ryn et al. 2000)[5]. Further, valdecoxib has been shown to cause substantially insignificant gastroduodenal ulcers than NSAIDs while the efficacy of the two groups is equal (Sikes et al. 2002)[6].

NSAIDs and coxibs are known to have multiple effects on kidney functions as seen in clinical trials. It has recently been suggested that even small elevations in blood pressure induced by NSAIDs could have a major effect on cardiovascular risk profile (Singh et al. 2003)[7].

Recently valdecoxib was withdrawn due to its atherosclerotic and cardiovascular adverse effects thus contraindicated in coronary artery disease (Soloman DH 2005)[8]. Sensing the remote relationship of the renal outcome on cardiac toxicity, the present study was conducted to observe the adverse outcome of diclo and valde in kidney of albino rats. The effects of the two drugs were compared and

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assessed along with biochemical changes at sub-therapeutic and therapeutic doses in rats.

MATERIAL AND METHODS

Animals

50 Adult, male & female Dukrey rats (180-200gm) in equal ratio were obtained from National Animals Laboratory Centre, Central Drug and Research Institute (CDRI), Lucknow. Experimental protocols were approved by Institutional Ethical Committee following the guideline of CPSCEA (Committee for the Purpose of Control and Supervision of Experiments on Animals) which competes with International norms of INSA (Indian National Science Academy).

Drugs

Diclofenac sodium and Valdecoxib were obtained from M/s Karnataka Antibiotic Pharmaceutical Ltd. and Glenmark Pharmaceuticals Ltd. respectively.

Enzyme Estimation

E-merk's diagnostic kits were used for the estimation of enzyme - serum creatine, BUN, uric acid, Na+ and K+ for renal function.

Dosage and administration

The doses administered in this study, were calculated according to the therapeutic dose as recommended for the chronic treatment of arthritis. We gave diclo at the doses of 5 and 10 mg/kg/day whereas valde at the doses of 1 and 2 mg/kg/day per orally. The subtherapeutic dose, half of the therapeutic dose was selected to see the level at which changes begins.

Methods

Rats of either sex were randomly divided into 5 groups of 10 each and drugs were administered per orally for 30 days as follows:

Group A - served as control was administered with normal saline only

Group B1 - Diclofenac sod. 5 mg / kg / day p.o.

Group B2 - Diclofenac sod. 10 mg / kg / day p.o.

Group C1 - Valecoxib 1 mg / kg /day p.o.

Group C2 - Valdecoxib 2 mg / kg/ day p.o.

Drugs solution was freshly prepared daily by dissolving in normal saline before oral administration. Blood sample - was collected after the end of treatment i.e. on day 30, from left ventricle of rats. Specimen collection - the control and treated group of animals were perfused with 10% formal saline. The kidneys were collected in 10% formalin and were processed for histopathology. The sections of the organs were stained using H/E (hematoxylin and eosin) and viewed under 40X magnification. Statistical analysis data of

Group (Drug/Dose mg/kg p.o.)	BUN (mmol/l)	S.Creatinine (µmol/l)	S.Uric acid (mmol/l)	S.Na⁺ (mmol/l)	S. K⁺ (mmol/l)
A(Control)	11.69± 1.59	32.26± 4.91	0.19± 0.06	146.00± 4.26	4.69± 0.40
B_1 (Diclofenac, 5)	11.84± 1.67	32.17± 5.39	0.21± 0.04	146.50± 3.02	6.62± 0.85***
B_2 (Diclofenac,10)	19.09± 2.45***	141.08± 40.60***	0.21± 0.05	143.80± 2.82	6.23± 0.91***
C_1 (Valdecoxib, 1)	19.28± 7.62**	79.55± 20.23***	0.43± 0.70	142.60± 2.41	6.46± 1.48**
C_2 (Valdecoxib, 2)	22.07± 3.88***	112.70± 39.49***	3.79± 1.09***	147.30± 4.27*	6.8± 0.08***

Table I: Results of renal biochemical parameters
The values are expressed as mean ± standard error.
* P<0.05, **P<0.01 *** P<0.001 as compared to control

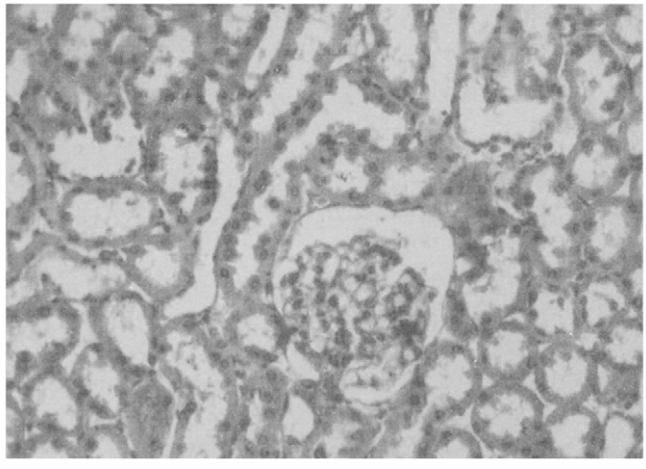

Fig 1a Kidney control group A (40X) showing cortex

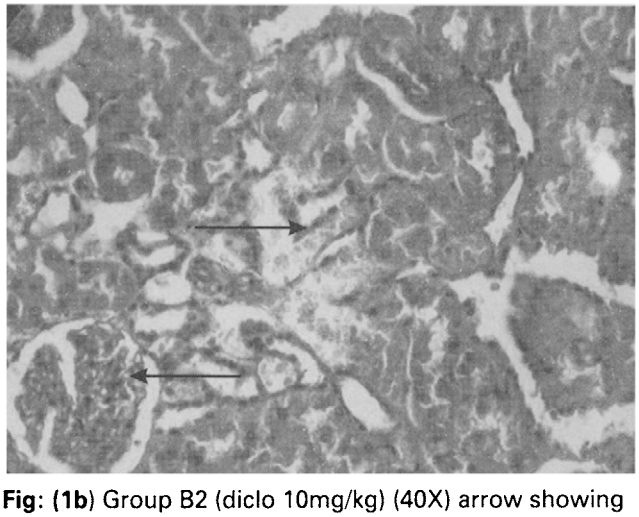

Fig: (1b) Group B2 (diclo 10mg/kg) (40X) arrow showing degenerating tubule

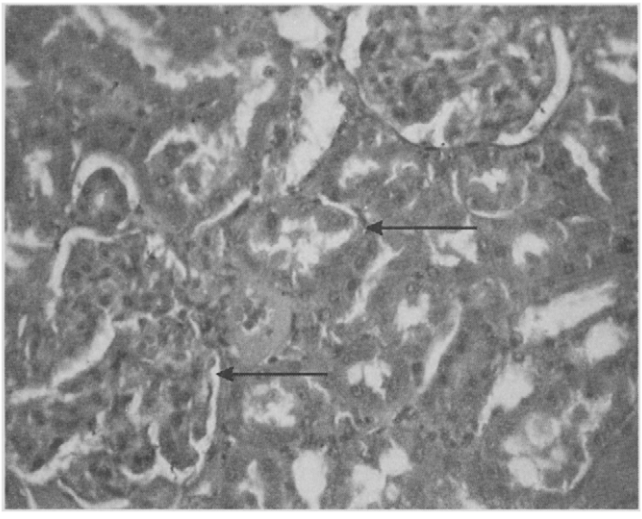

Fig: (1c) Group C1 (valde 1mg/kg) (40X) Large arrow showing increase cellularity of glomerulus & small arrow showing degenerating tubule

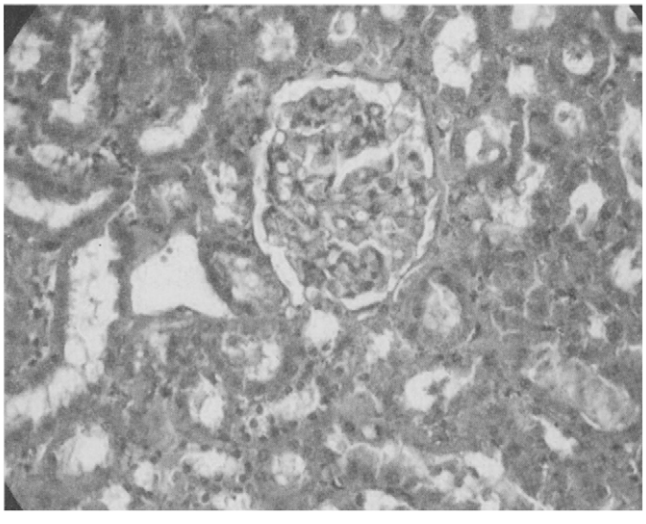

Fig: (1d) Group C2 (valde 2mg/kg) (40X) more marked feature than group C1

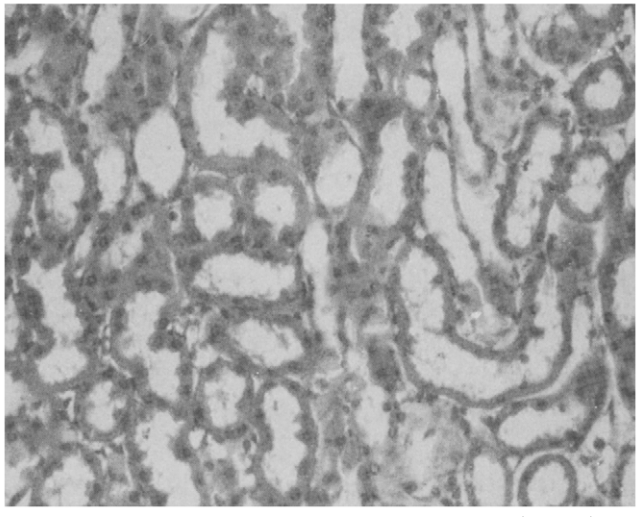

Fig: (2a) Kidney control group A (40X) showing medulla

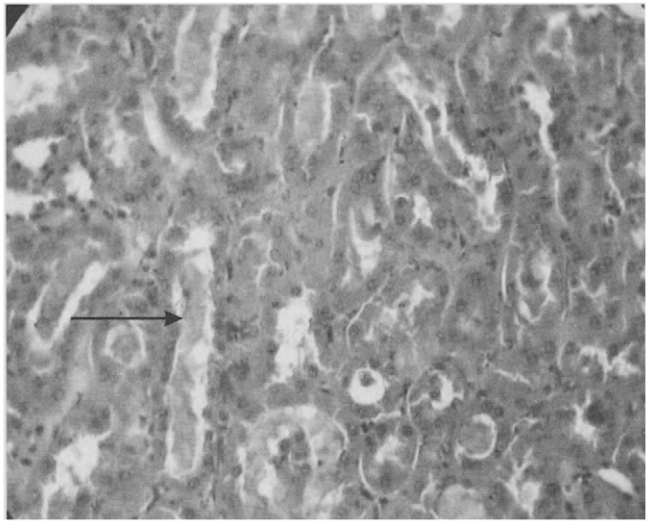

Fig: (2b) Group B2 (diclo 10mg/kg) arrow showing degenerating tubule with intraluminal exudates

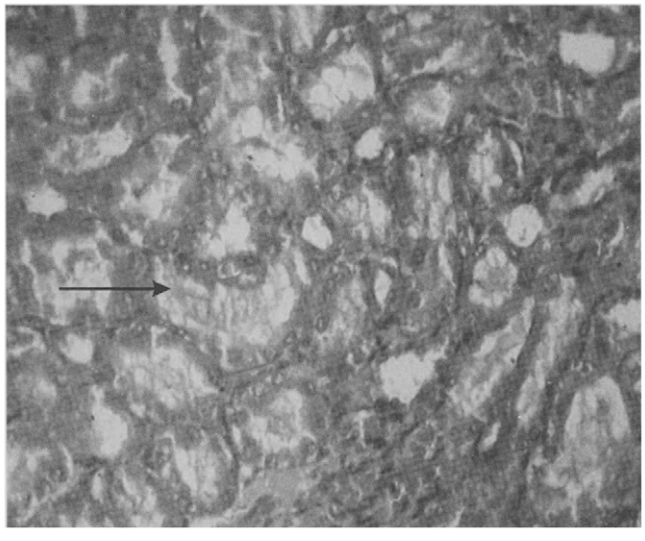

Fig: (2c) Group C1 (Valde 1mg/kg)(40x) arrow showing degenerating tubule with intraluminal protein exudates

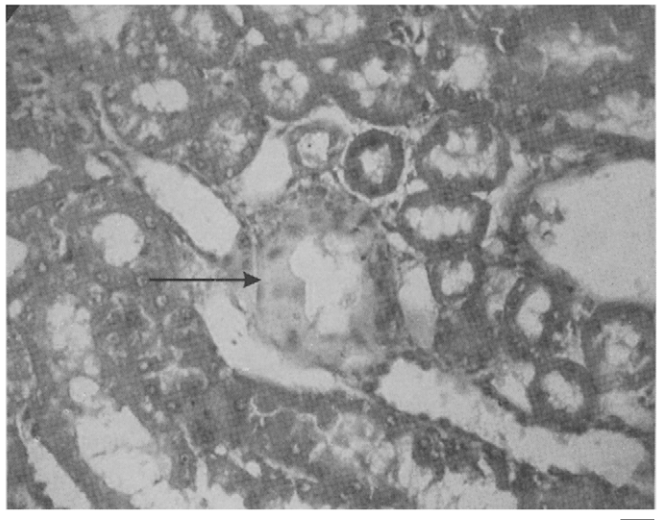

Fig: (2d) Group C2 (valde 2mg/kg) (40X) arrow showing degenerating tubule with cloudy swelling

biochemical parameters were presented as mean + SEM (Standard Error of Mean) for control and experimental animals.

RESULTS

Histological changes

The kidney sections of Group B1 showed no significant change when compared with the control Group A (fig: 1a). Group C1 showed degenerative and denudative changes in the epithelium of proximal and distal convoluted tubule (fig: 1c). Tubules in medulla showed degenerative changes with intraluminal protein exudates (fig: 2c) in comparison to control group A (fig: 2a). Group B2 showed increase in cellularity of glomerulus with mild thickening of glomerular basement membrane. Degenerative changes were observed in proximal and distal convoluted tubules (fig: 1b). Tubule in medulla showed degenerative changes with intraluminal exudates (fig: 2b). Group C2 showed increased cellularity of glomerulus with thickening of glomerular basement membrane along with degenerative changes. Proximal tubule showed marked degenerative changes and vacuolation (fig: 1d). Medulla showed degenerating tubules with intraluminal protein exudates in abundance. Dilated vessels were seen scattered in the interstitium (fig: 2d)

Biochemical changes: - creatine showed significant increase in groups B2, C1 and C2 ($p<0.001$). BUN showed significantly increased levels in groups B2 & C2 ($p<0.001$) and C1 ($p<0.01$). UA was found to be significantly raised in group C2 ($p<0.001$). Na+ was observed to be significantly increased in group C2 ($p<0.05$) only. K+ was significantly increased in group B1, B2&C2 ($p<0.001$) and with C1 ($p<0.01$) (Table- I).

DISCUSSION

Selective COX-2 inhibitor is proposed to be as effective as traditional NSAIDs but would spare the gastrointestinal mucosa (Coruzzi et al 2004)[9]. Valdecoxib was banned due to atherosclerosis and cardiovascular effects (Soloman DH 2005)[8]. Studies are lacking in correlating the link between the renal dysfunction to cardiac disease. Fluid overload may lead to congestive heart failure, accumulation of urea can lead to pericarditis and cardiac arrythmias can be associated to potassium accumulation (Perazella & Khan 2006)[10]. Chronic renal failure patients suffer from accelerated atherosclerosis and high incidence of cardiovascular disease (Hojs R et al. 2002)[11]. Earlier studies with non-selective NSAIDs, naproxen by Schwatz et al. (2002)[12] showed similar result as observed on renal histological examination with raised biochemical estimation and electrolyte level. Similar results were to present study observed by Harifarrosh and Jamali (2005)[13] with diclofenac and flurbiprofen. Prakashreddy et al. (2006)[14] reported same changes with diclofenac just as Segal et al. (2006)[15] observed with aspirin.

In the present study, findings using higher dose of diclofenac is in accordance with the observations in all these prior studies. But use of low dose diclofenac showed normal histology and biochemical parameters, only serum potassium was increased. Earlier observations with coxibs showed variable results, toxic outcome was seen in study of the Schattener et al. (2000)[16], Patricia et al. (2002)[17] with nimesulide, Schwatz et al (2002)[12] and Harifarrosh and Jamali(2005)[13]with rofecoxib and celecoxib. Present study, showing adverse effects with both the doses of valdecoxib, thereby supports the above studies but contradicts those studies, which predict safe profile as showed with nimesulide by Barskova et al. (2004)[18] and Prakashreddy et al. (2006)[14].

The results of the present study add to distinct heterogeneity within widely prescribed class of drugs. Thus, sub-therapeutic and therapeutic doses of valdecoxib, on long-term exposure can lead to cardiac problems i.e. CHF, arrythmias and atherosclerosis. Whereas with diclofenac, it therapeutic dose can lead to pathological changes while its sub therapeutic dose seems to be safer than valdecoxib in renal outcomes. Thus, it can be inferred that changes begin at subtherapeutic level for COX-2 inhibitor. Therefore these drugs need to be taken with precaution in patient of renal compromised conditions.

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