RESULTS

1. Effect of dehydration and glycerol (10 mg/kg I.M) on kidney function in male albino rats:

Table (1) shows that dehydration and glycerol (10 mg/kg I.M) cause significant ($P < 0.01$) increase in serum urea (5.11 ± 1.07 to 19.25 ± 1.43 and 10.51 ± 1.8), serum creatinine (56.675 ± 7.32 to 200.92 ± 28.83 and 91.6 ± 8.66), serum potassium (5.46 ± 0.936 to 16.9 ± 0.786 and 22.78 ± 0.72), serum calcium (0.64 ± 0.119 to 2.94 ± 0.158 and $2.48 \pm - 0.21$) and serum sodium (133.18 ± 11.7 to 184.43 ± 11.72 and 196.49 ± 17.072) in rats sacrificed 2 days and 7 days after glycerol injection respectively (Table 2).

2. Effect of diltiazem (9 mg/kg orally twice/day) started 7 days before dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 2 days after dehydration in comparison with normal control group and prophylactic control group sacrificed 2 days after dehydration.

Table (2) show insignificant difference ($P > 0.05$) in serum urea (5.11 ± 1.70 & 5.82 ± 0.49 with +14% change), serum creatinine (56.675 ± 7.32 & 45.447 ± 3.22 with -20% change), serum potassium (5.46 ± 0.936 & 3.56 ± 0.323 with -35% change) and serum sodium (133.18 ± 11.7 & 132.76 ± 6.35 with -1% change) between the normal control group and the prophylactically diltiazem treated group respectively. The same table shows significant ($P < 0.05$) increase in serum calcium in prophylactically diltiazem treated uraemic group (1.35 ± 0.182) than that observed in normal control group (0.64 ± 0.119). Also, this table shows statistically significant difference (P < 0.05) in serum urea (19.25 ± 1.43 to 5.82 ± 0.49 with -70% change), serum creatinine (200.92 ± 28.83 to 45.447 ± 3.22 with -77% change), serum potassium (16.9 ± 0.786 to 3.65 ± 0.323 with -78% change), serum calcium (2.94 ± 0.158 to 1.35 ± 0.182 with -54% change) and serum sodium (184.43 ± 11.72 to 132.76 ± 6.35 with -28% change) (Table 2).

3. Effect of diltiazem (9 mg/kg twice daily orally) started 2 days after dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 9 days after dehydration compared to normal control group and curative control uraemic group sacrificed 9 days after dehydration.

Table (3) shows insignificant difference P > 0.05) in serum urea (5.11 ± 1.07 & 3.73 ± 0.78 with -27% change), serum creatinine (56.675 ± 7.32 & 62.46 ± 6.86 with 1% change) and serum sodium (133 ± 11.72 & 129.4 ± 11.71 with -3% change) while shows significant difference (P < 0.05) in serum potassium (5.46 ± 0.936 & 21.49 ± 1.296 with +249% change) and serum calcium (0.64 ± 0.119 & 1.57 ± 0.087 with 145% change) between normal control group and diltiazem curatively treated uraemic group. Also, this table shows significant difference (P < 0.05) in serum urea (10.51 ± 1.18 to 3.73 ± 0.78 with -65% change), serum creatinine (91.6 ± 8.66 to 62.46 ± 6.86 with -32% change), serum calcium (2.48 ± 0.21 to 1.57 ± 0.087 with -

36% change), and serum sodium 196.49 ± 17.072 to 129.4 ± 11.71 with 34% change) while shows insignificant difference ($P > 0.05$) in serum potassium (22.78 ± 0.72 to 21.49 ± 1.296 with -6% change) between the curative Diltiazem treated uraemic rats and the curative untreated control uraemic group (Table 3).

4. Effect of benazepril (31 mg/kg/day orally) started 7 days before dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 2 days after dehydration in comparison with normal control group and control prophylactic uraemic group sacrificed 2 days after dehydration

Table (4) shows statistically significant difference ($P < 0.01$) in serum urea (5.11 ± 1.07 & 17.53 ± 1.06 with 243% change), serum creatinine (56.675 ± 7.32 & 207.9 ± 37.18 with 267% change), serum potassium (5.46 ± 0.936 & 10.2 ± 0.52 with 87% change), serum calcium (0.64 ± 0.119 & 2.972 ± 0.352 with 365% change) and serum sodium (133.18 ± 11.7 & 200.4 ± 14.135) between the normal control group and benazepril prophylactically treated uraemic group. Also, this table shows insignificant difference ($P > 0.05$) in serum urea (19.25 ± b1.43 to 17.53 ± 1.06 with -9% change), serum creatinine (200.92 ± 28.83 to 207.9 ± 37.18 with 3% change), serum potassium (16.9 ± 0.786 to 10.2 ± 0.52 with -40% change), serum calcium (2.94 ± 0.158 to 2.975 ± 0.352 with +1% change) and serum sodium 184.43 ± 11.72 to 200.4 ± 14.135 with +9% change) between control prophylactic uraemic group and benazepril prophylactically treated group (Table 4).

5. Effect of benazepril (31 mg/kg/day orally) started 2 days after dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 9 days after dehydration in comparison with normal control group and curative control uraemic group sacrificed 9 days after dehydration.

Table (5) show statistically significant difference ($P < 0.05$) on serum urea (5.11 ± 1.07 & 8.67 ± 1.16 with +70% change), serum creatinine (56.675 ± 7.32 & 114.05 ± 6.287 with +101% change), serum potassium (5.46 ± 0.936 & 21.8 ± 0.837 with +299% change), serum calcium (0.64 ± 0.119 & 2.69 ± 0.212 with +320% change) and serum sodium (133 ± 11.7 & 193.7 ± 13.42 with +46% change) between normal control group and benazepril curatively treated uraemic group. Also, this table show insignificant difference ($P > 0.05$) in serum urea (10.51 ± 1.18 to 8.67 ± 1.16 with -18% change), serum creatinine (91.6 ± 8.66 to 114.05 ± 6.287 with +25% change), serum potassium (22.78 ± 0.72 to 21.8 ± 0.837 with + 8% change), serum calcium (2.48 ± 0.21 to 2.69 ± 0.212 with -4% change) and serum sodium (196.49 ± 17.072 to 193.7 ± 13.42 with -1% change) between the benazepril curatively treated uraemic group and the curative control uraemic group (Table 5).

Histopathological results:

1) Normal control group:

The kidney show an outer cortex and an inner striated-appearing medulla. The renal medulla was composed of conical medullary pyramides, whose bases form the corticomedullary junction and whose vertices protrude into the minor calyxes. From the bases of each pyramid, parallel medullary rays penetrate the cortex, each was consisting of one or more collecting tubules together with the straight portion of several nephrons surrounding each medullary ray. A cortical labyrinth was found consisting of renal corpuscles and the convoluted portions of the nephron. The renal corpuscles appeared formed of tuft of capillaries, glomeruli, surrounded by double walled epithelial corpuscle, the Bowman's capsule. Closely related to renal corpuscle, there was many section of the convoluted tubules. Two types were evident, the first one is representation of proximal convulsion. The cells of the proximal convulsion were of simple cuboidal type, having an acidophilic granular cytoplasm containing spherical nuclei, usually located in the center, and the cell apex possessed a characteristic irregular brush border as an out standing morphologic features of these cells was the absence of distinct cell boundaries. The second type of convolution was the distal convulsion. Which was characterized by flat smaller cells which were less acidophilic, did not have a prominent brush border with more or less distinct cell boundaries (Fig. 2).

2) Dehydration and Glycerol prophylactic control group (GPC):

There were a serious structural changes primarily affecting the epithelium of the proximal convoluted tubules. The changes seem to be that type called tubular degeneration evidenced by the presence of apparent cytoplasmic vacuoles. Most of the affected tubules were dilated and filled homogenous hyalinised casts (Fig. 3).

3) Dehydration and Glycerol curative control group (Gcc):

There were more tubular damage than that observed in GPc. The damaged tubular epithelium was sloughed with subsequent loss of its integrity (necrosis) and many tubules appeared full of cellular debris (Fig. 4).

4) Effect of prophylactic diltiazem treatment on dehydration and glycerol induced renal changes:

Diltiazem starting 7 days before dehydration abolishes the glycerol induced structural changes to the extent that the tubular epithelium of proximal convoluted tubules regained their normal morphological characteristics. The tubules appeared lined with intact truncated cells with their apparent cytoplasmic acidophilic and their spheroidal nuclei. The luminal borders of these cells appeared to be provided with obvious brush border (Fig. 5).

5) Effect of curative diltiazem treatment on glycerol induced renal changes:

Diltiazem given 2 days after dehydration for 7 days moderately improved the epithelium of proximal convoluted tubules which exhibited moderate degenerative changes without necrosis.

6) Effect of prophylactic benazepril on dehydration and glycerol induced renal changes:

Benazepril given 7 days before dehydration a meliorated to small extent the degenerative changes induced by glycerol.

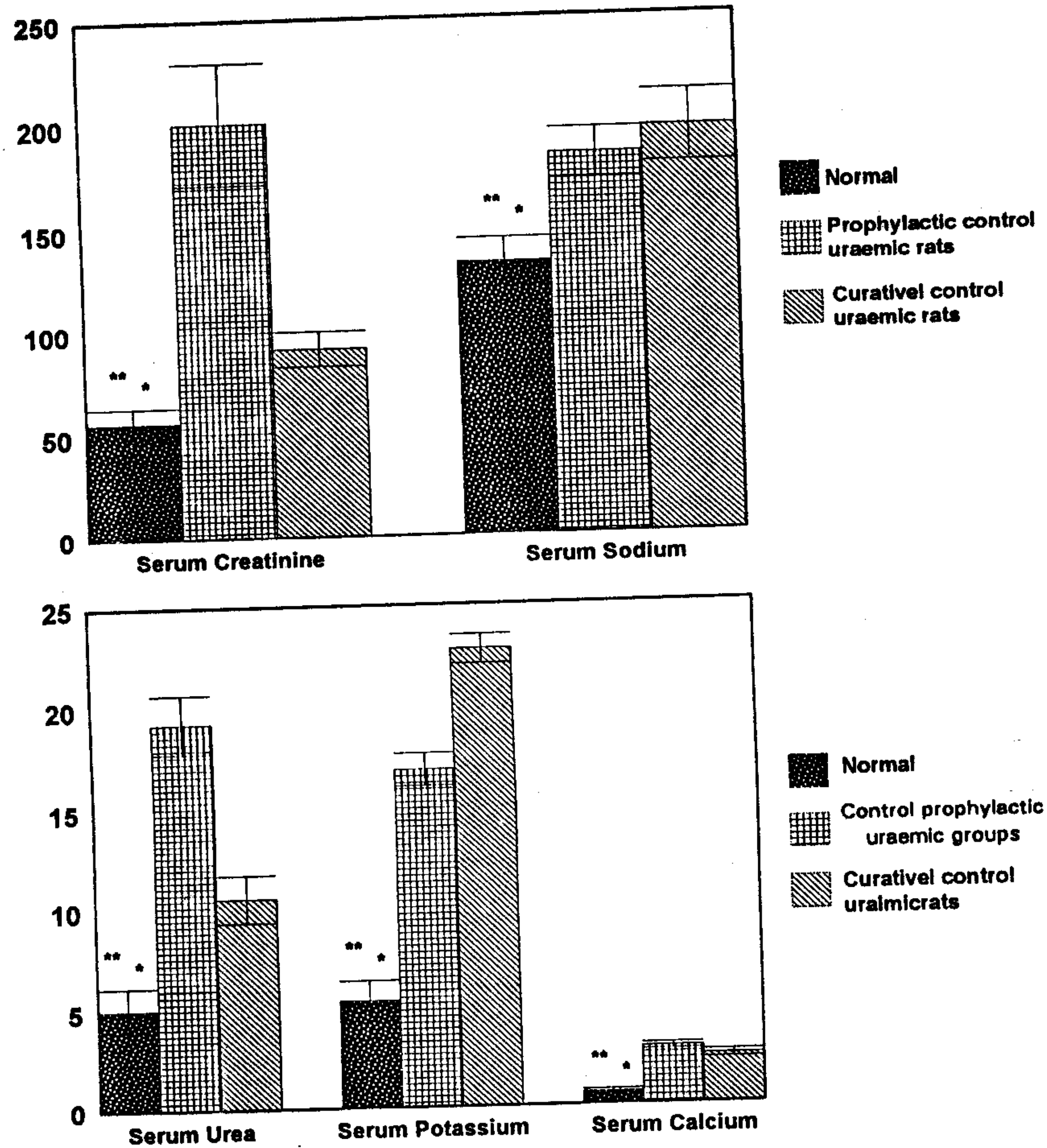
7) Effect of curative benazepril on glycerol induced renal change:

Benazepril given 2 days after dehydration for 7 days did not exert a marked effect on the previously occurred structural changes as the examined specimen showed a persistence of nearly the same degree of tubular damage.

Table (1) : Effect of dehydration and glycerol (10 mg/kg I.M) on kidney functions of rats.

Kidney function Parameters	Normal	dehydration and Glycerol induced uraemic rats	
		Sacrified 2dayes after dehydration	Sacrified 9 dayes after dehydration
Serum Urea	(n=8)	(n=9)	(n=8)
Mean (M mol/L)	5.11	19.25	10.51
± S.E	± 1.07	± 1.43	± 1.18
P		P< 0.01	P< 0.01
% change		+ 277 %	+ 106 %
Serum Creatinine	(n=10)	(n=10)	(n=10)
Mean (M mol/L)	56.675	200.92	91.6
± S.E	± 7.32	± 28.83	± 8.66
P		P< 0.01	P< 0.01
% change		+ 255 %	+ 67 %
Serum Potassium	(n=10)	(n=8)	(n=8)
Mean (M mol/L)	5.46	16.9	22.78
± S.E	± 0.936	± 0.786	± 0.72
P		P< 0.01	P< 0.01
% change		+ 210 %	+ 317 %
Serum Calcium	(n=12)	(n=8)	(n=8)
Mean (M mol/L)	0.64	2.94	2.48
± S.E	± 0.119	± 0.158	± 0.21
P		P< 0.01	P< 0.01
% change		+ 359 %	+ 288 %
Serum Sodium	(n=7)	(n=9)	(n=8)
Mean (M mol/L)	133.18	184.43	196.49
± S.E	± 11.7	± 11.72	± 17.072
P		P< 0.01	P< 0.01
% change		+ 116 %	+ 48 %

$P < 0.01$ means significant difference.
N = number of readings.



* Significant (P< 0.01) compared with prophylactic control uraemic rats.

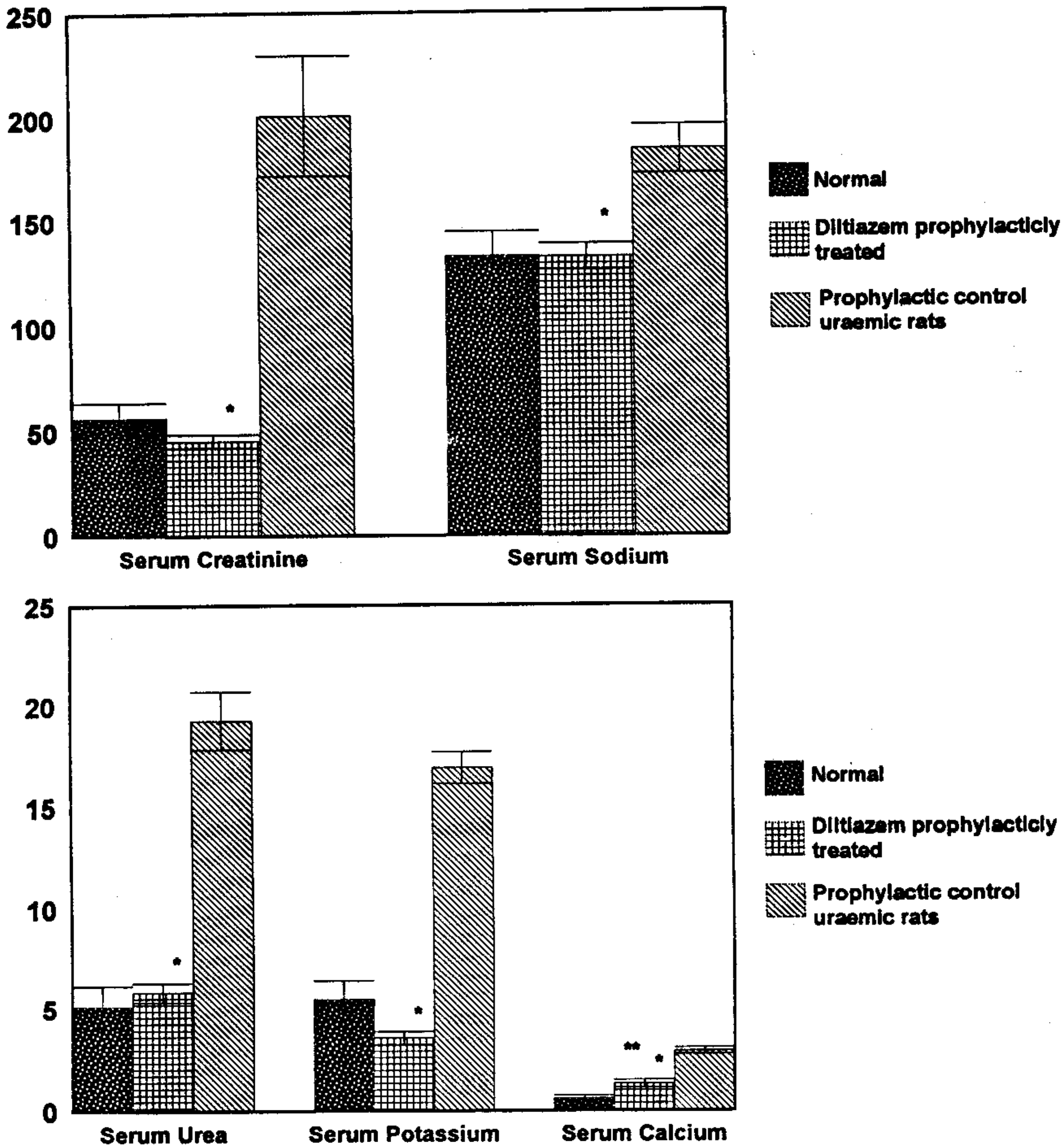
** Significant (P< 0.01) compared with curative control uraemic rats.

Fig.(2): Bar chart showes the effect of dehydration and glycerol (10 mg/kg I.M) on kidney functions of rats.

Table (2) : Effect of diltiazem (9 mg/kg twice daily orally) started 7 dayes befor dehydration on kidney functions of dehydration and glyceral induced uraemic rats sacrificd 2 dayes after dehydration in comparison with the normal group, and control prophylactic uraemic group sacrified 2 dayes after dehydration.

Kidney function	Normal	Diltiazem treated (prophylactic) glycerol induced uraemic rats	Control prophylactic uraemic groups
Serum Urea Mean (M mol/L) ± S.E P % change	(n=8) 5.11 ± 1.07	(n=6) 5.82 ± 0.49 P> 0.05 (vs Normal) + 14 %	(n=9) 19.25 ± 1.43 P< 0.05 (vs Diltiazem treated) - 70 %
Serum Creatinine Mean (M mol/L) ± S.E P % change	(n=10) 56.675 ± 7.32	(n=10) 45.447 ± 3.22 P> 0.05 (vs Normal) - 20 %	(n=10) 200.92 ± 28.83 P< 0.05 (vs Diltiazem treated) - 77 %
Serum Potassium Mean (M mol/L) ± S.E P % change	(n=10) 5.46 ± 0.936	(n=11) 3.56 ± 0.323 P> 0.05 (vs Normal) - 35 %	(n=8) 16.9 ± 0.786 P< 0.05 (vs Diltiazem treated) - 78 %
Serum Calcium Mean (M mol/L) ± S.E P % change	(n=12) 0.64 ± 0.119	(n=8) 1.35 ± 0.182 P< 0.05 (vs Normal) + 11 %	(n=8) 2.94 ± 0.168 P< 0.05 (vs Diltiazem treated) - 54 %
Serum Sodium Mean (M mol/L) ± S.E P % change	(n=7) 133.18 ± 11.7	(n=9) 132.76 ± 6.35 P> 0.05 (vs Normal) - 1 %	(n=9) 184.43 ± 11.72 P< 0.05 (vs Diltiazem treated) - 28 %

P > 0.05 means insignificant difference.
P < 0.05 means significant difference.



* Significant (P< 0.05) compared with prophylactic control uraemic rats.

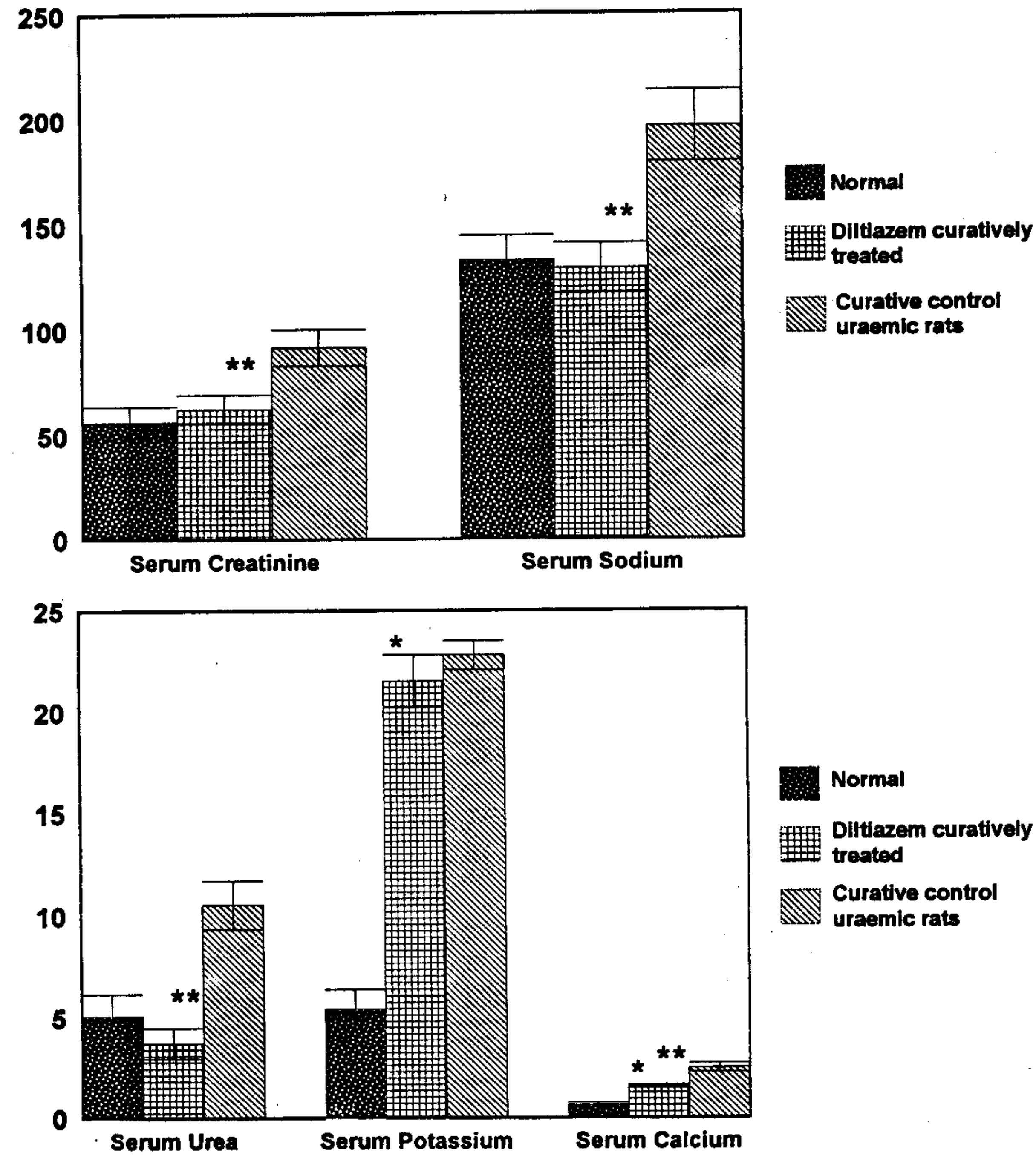
** Significant (P< 0.05) compared with normal rats.

Fig.(3): Bar chart showes the effect of diltiazem (9 mg/kg twice daily orally) started 7 days befor dehydration on kidney functions of dehydration and glyceral induced uraemic rats sacrificed 2 days after dehydration in comparison with the normal group and curative control uraemic group sacrificed 2 days after dehydration.

Table (3) : Effect of diltiazem (9 mg/kg twice daily orally) started 2 dayes after dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificd 9 dayes after dehydration in comparison with the normal group, and curative control uraemic group sacrified 9 days after dehydration.

Kidney function	Normal	Diltiazem + treated (curative) glycerol induced uraemic rats	curative control uraemic group sacrified 9 dayes after dehydration.
Serum Urea Mean (M mol/L) ± S.E P % change	(n=8) 5.11 ± 1.07	(n=9) 3.73 ± 0.78 P> 0.05 - 27 %	(n=8) 10.51 ± 1.18 P< 0.05 - 65 %
Serum Creatinine Mean (M mol/L) ± S.E P % change	(n=10) 56.675 ± 7.32	(n=9) 26.46 ± 6.86 P> 0.05 - 1 %	(n=10) 91.6 ± 8.66 P< 0.05 - 32 %
Serum Potassium Mean (M Eq/L) ± S.E P % change	(n=10) 5.46 ± 0.936	(n=10) 21.49 ± 1.296 P< 0.05 + 249 %	(n=8) 22.78 ± 0.72 P> 0.05 - 6 %
Serum Calcium Mean (M mol/L) ± S.E P % change	(n=12) 0.64 ± 0.119	(n=8) 1.57 ± 0.087 P< 0.05 + 145 %	(n=8) 2.48 ± 0.21 P< 0.05 - 36 %
Serum Sodium Mean (M Eq/L) ± S.E P % change	(n=7) 133.18 ± 11.7	(n=6) 129.4 ± 11.71 P> 0.05 - 3 %	(n=8) 196.49 ± 17.072 P< 0.05 - 34 %

P > 0.05 means insignificant difference.
P < 0.05 means significant difference.



* Significant (P< 0.05) compared with normal group

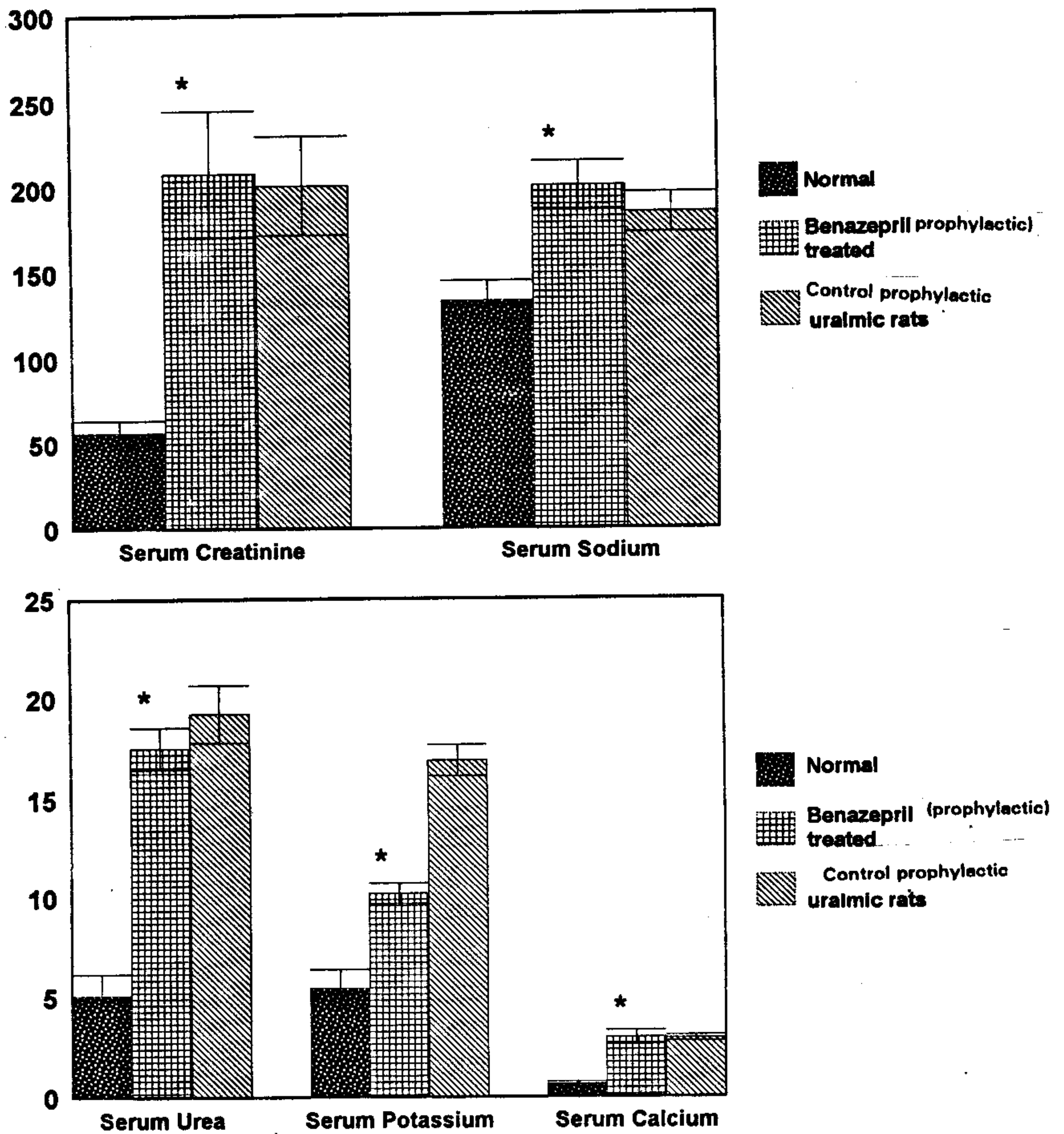
** " " " " curative control uraemic rats.

Fig.(4): Bar chart showes the effect of diltiazem (9 mg/kg twice daily orally) started 2 days after dehydration on kidney functions of dehydration and glyceral induced uraemic rats sacrificed 9 days after dehydration in comparison with the normal group and prophylactic control uraemic group sacrificed 9 days after dehydration.

Table (4) : Effect of benazepril (1 mg/kg once daily orally) started 7 dayes before dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 2 dayes after dehydration in comparison with the normal group, and curative control uraemic group sacrified 2 dayes after dehydration.

Kidney function	Normal	Benazepril + treated (prophylactic) glycerol induced uraemic rats	Control prophylactic uraemic group
Serum Urea	(n=8)	(n=8)	(n=8)
Mean (M mol/L)	5.11	17.53	19.25
± S.E	± 1.07	± 1.06	± 1.43
P		$P < 0.01$	$P > 0.05$
% change		+ 243 %	- 9 %
Serum Creatinine	(n=10)	(n=7)	(n=10)
Mean (M mol/L)	56.675	207.9	200.92
± S.E	± 7.32	± 37.18	± 28.83
P		$P < 0.01$	$P > 0.05$
% change		+ 267 %	+ 3 %
Serum Potassium	(n=10)	(n=8)	(n=8)
Mean (M mol/L)	5.46	10.2	16.9
± S.E	± 0.936	± 0.52	± 0.786
P		$P < 0.01$	$P > 0.05$
% change		+ 87 %	- 40 %
Serum Calcium	(n=12)	(n=8)	(n=8)
Mean (M mol/L)	0.64	2.975	2.94
± S.E	± 0.119	± 0.352	± 0.158
P		$P < 0.01$	$P > 0.05$
% change		+ 365 %	+ 1 %
Serum Sodium	(n=7)	(n=7)	(n=9)
Mean (M mol/L)	133.18	200.4	184.43
± S.E	± 11.72	± 14.135	± 11.72
P		$P < 0.01$	$P > 0.05$
% change		+ 50 %	+ 9 %

$P < 0.01$ means significant difference.
$P > 0.05$ means insignificant difference.



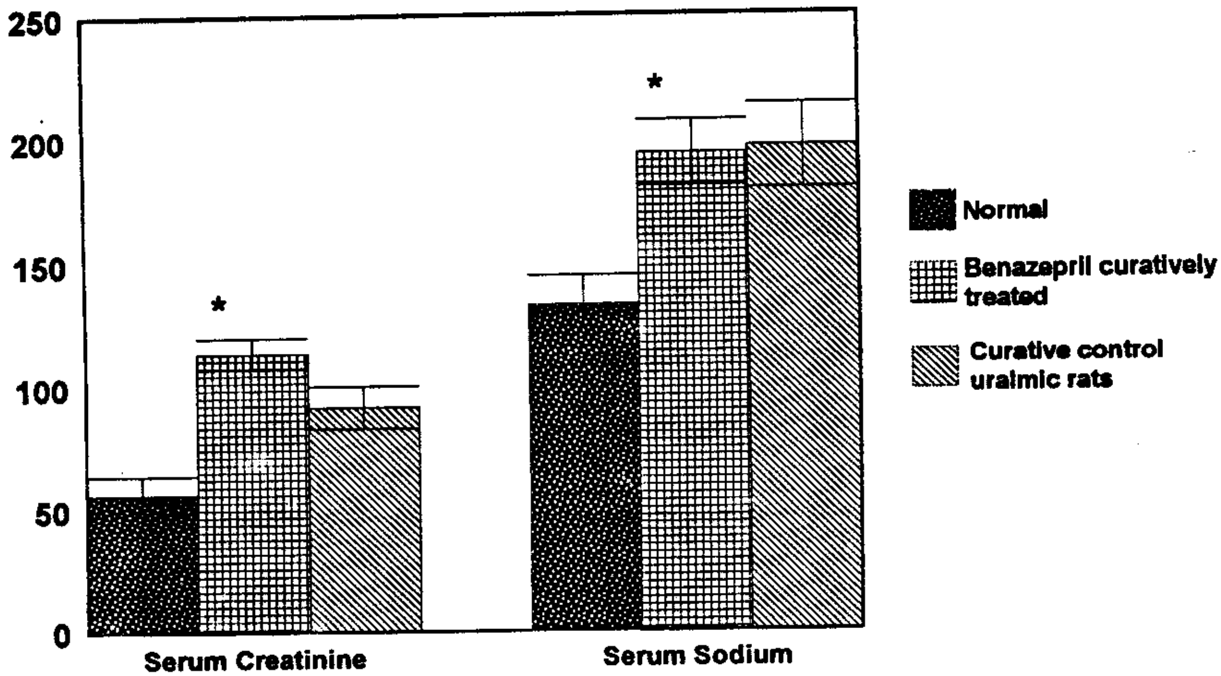
* Significant (P< 0.01) compared with normal group

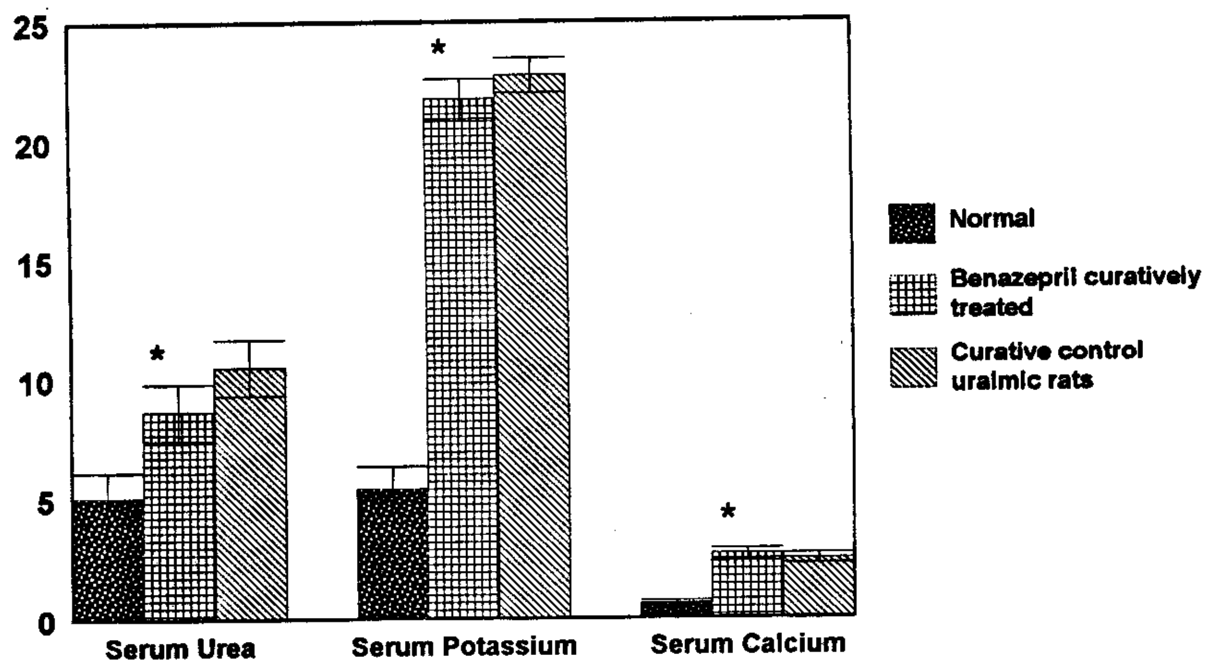
Fig.(5): Bar chart showes the effect of benazepril (31 mg/kg daily orally) started 7 days before dehydration on kidney functions of dehydration and glyceral induced uraemic rats sacrificed 2 days after dehydration in comparison with the normal group and prophylactic control uraemic group sacrificed 2 days after dehydration.

Table (5) : Effect of Benazepril(31 mg/kg once daily orally) started 2 dayes after dehydration on kidney functions of dehydration and glyceral induced uraemic rats sacrificed 9 dayes after dehydration in comparison with the normal group, and curative control uraemic group sacrified 9 dayes after dehydration.

Kidney function	Normal	Benazepril + treated (curative) glycerol induced uraemic rats	curative control uraemic group sacrified 9 dayes after dehydration.
Serum Urea	(n=8)	(n=6)	(n=8)
Mean (M mol/L)	5.11	8.67	10.51
± S.E	± 1.07	± 1.16	± 1.18
P		$P < 0.05$	$P > 0.05$
% change		+ 70 %	- 18 %
Serum Creatinine	(n=10)	(n=7)	(n=10)
Mean (M mol/L)	56.675	114.05	91.6
± S.E	± 7.32	± 6.287	± 8.66
P		$P < 0.01$	$P > 0.05$
% change		+ 101 %	+ 25 %
Serum Potassium	(n=10)	(n=8)	(n=8)
Mean (M mol/L)	5.46	21.8	22.78
± S.E	± 0.936	± 0.837	± 0.72
P		$P < 0.01$	$P > 0.05$
% change		+ 299 %	+ 8 %
Serum Calcium	(n=12)	(n=8)	(n=8)
Mean (M mol/L)	0.64	2.69	2.48
± S.E	± 0.119	± 0.212	± 0.21
P		$P < 0.01$	$P > 0.05$
% change		+ 320 %	- 4 %
Serum Sodium	(n=7)	(n=8)	(n=8)
Mean (M mol/L)	133.18	193.7	196.49
± S.E	± 11.72	± 13.42	± 17.072
P		$P < 0.01$	$P > 0.05$
% change		+ 46 %	- 1 %

$P < 0.05$ means significant difference.
$P > 0.05$ means insignificant difference.





* Significant (P< 0.05 or 0.01) compared with normal group

Fig.(6): Bar chart showes the effect of benazepril (31 mg/kg daily orally) started 2 days after dehydration on kidney functions of dehydration and glycerol induced uraemic rats sacrificed 9 days after dehydration in comparison with the normal group and prophylactic control uraemic group sacrificed 9 days after dehydration.

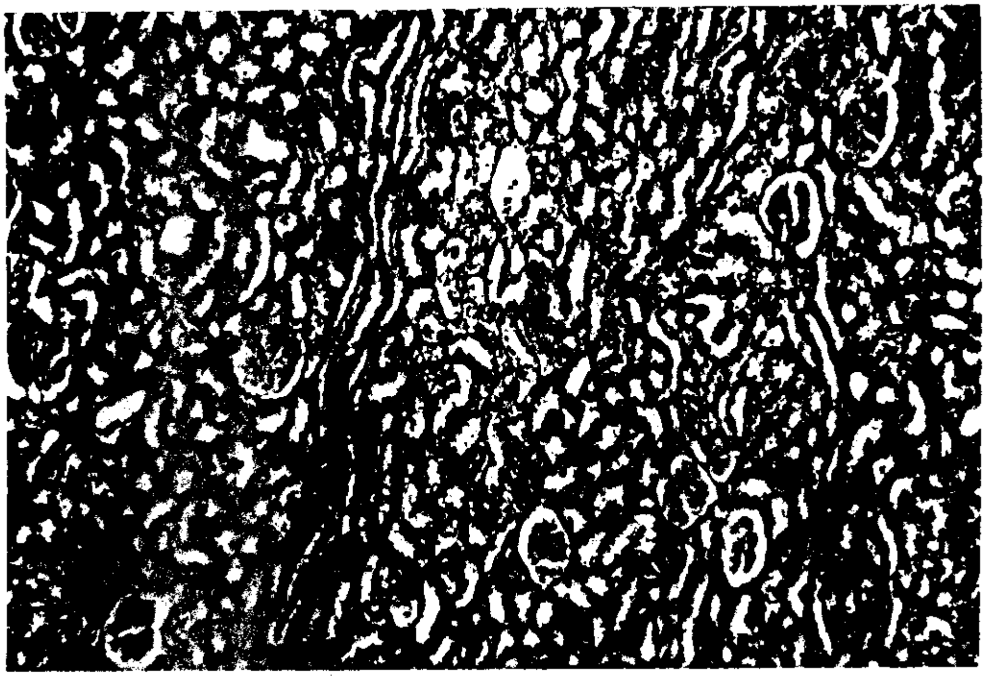


Fig (7) Photomicrograph of portion of renal cortex of normal control rat stained with H&Estain (Ax 125, BX 250 and CX 640).

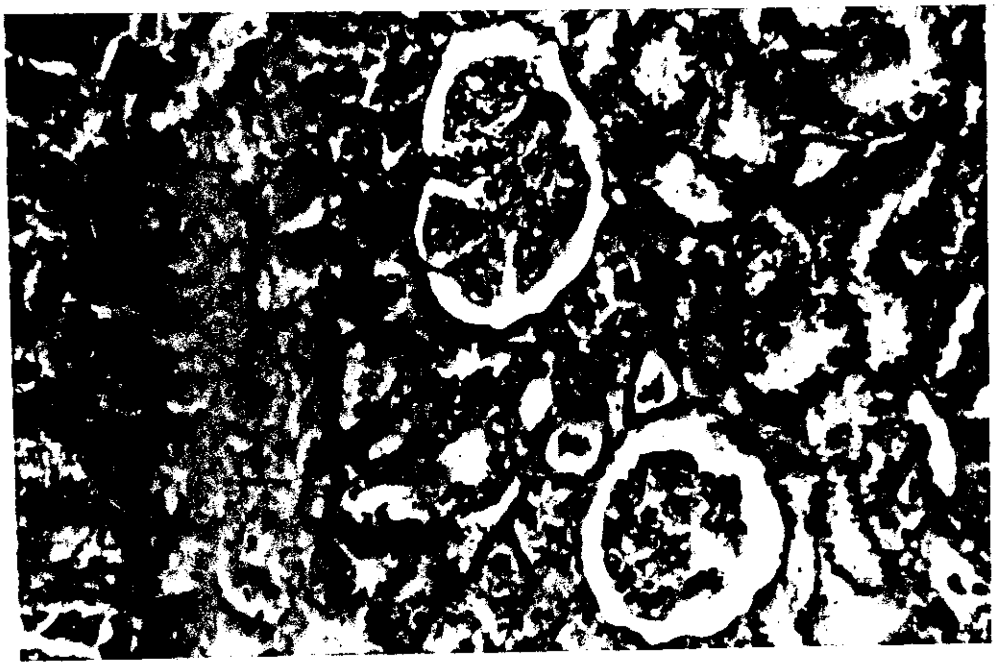


Fig (8) Photomicrograph of portion of renal cortex of Prophylatic control uralmic rats stained with H&Estaim shows degeneration of proximal convaluted tubular cells . (X250)

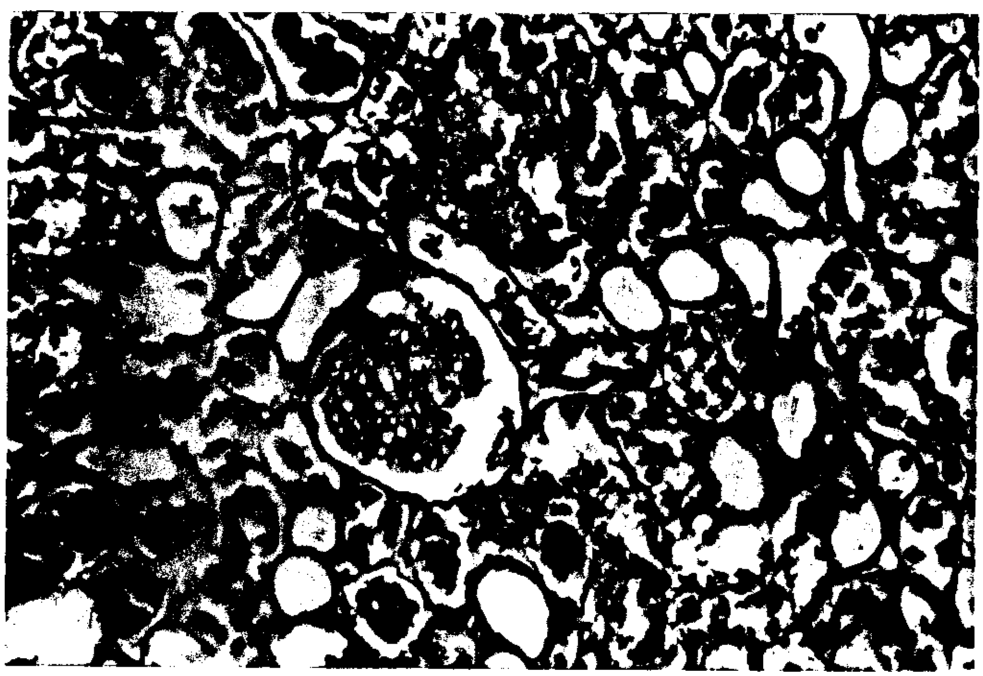


Fig (9) Photomicrograph of portion of renal cortex of cutative control uraemic rats stained with H&E stain shows nectosis of proximal tubular cells and cellular debris in its lumen (X 250)

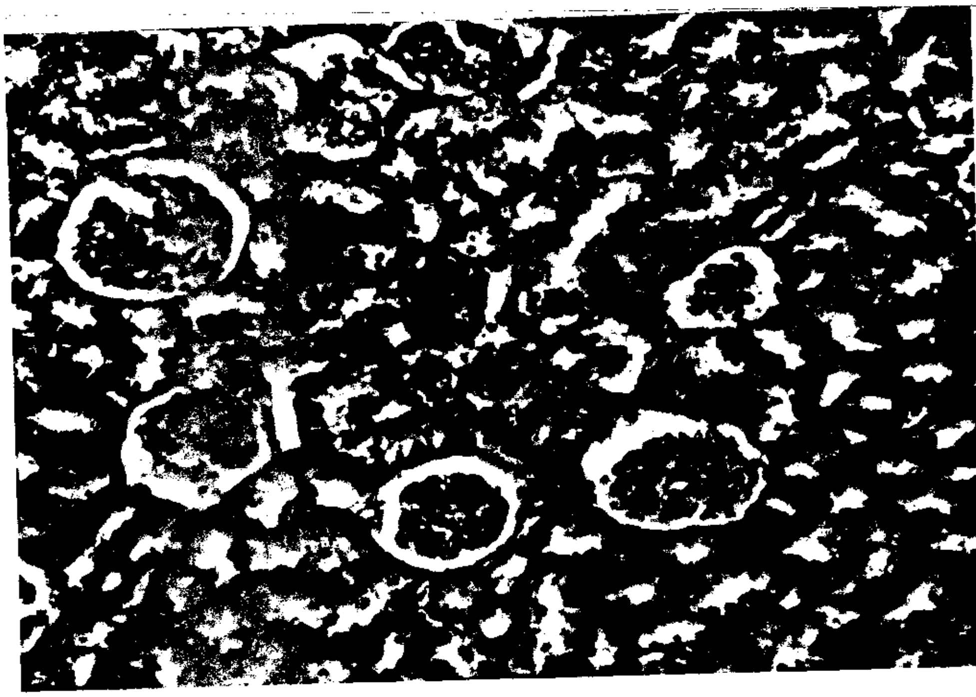


Fig (10) Photomicrograph of portion of renal cortex of prophylactic diltiazem treated uraemic rats stained with H&E stain shows nearly normal appearance (X 250).

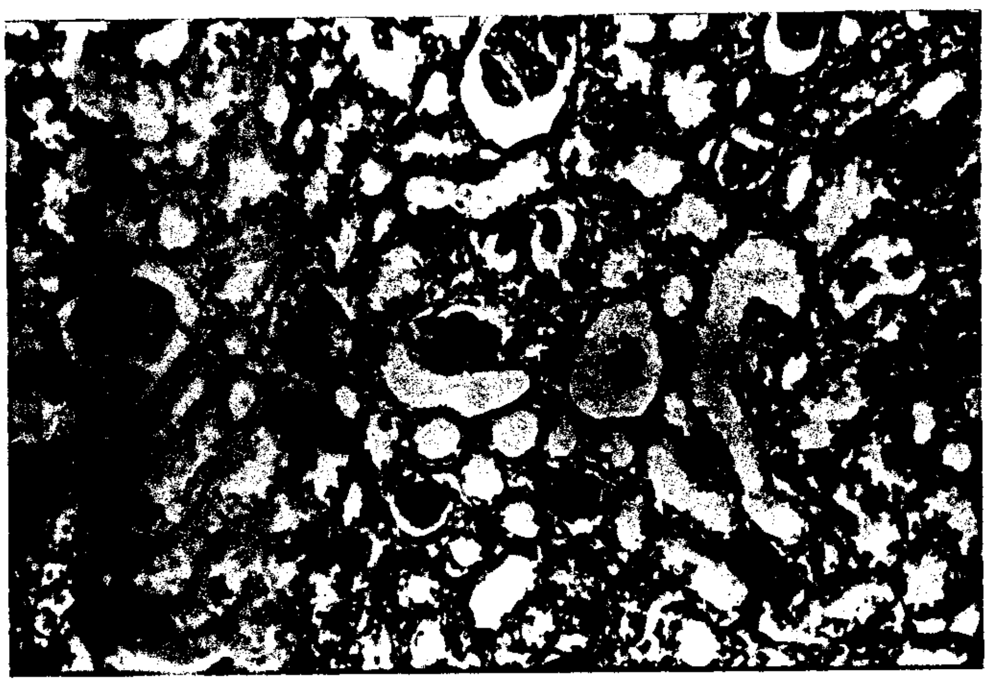


Fig (11) Photomicrograph of portion of renal cortex of curative diltiazem treated uraemic rats stained with H&E stain shows moderate degenerative changes in proximal convoluted tubular cells without necrosis (X 250).

Fig (12) Photomicrograph of portion of renal cortex of prophylactic benazepril treated uraemic rats stained with H&E stain shows degenerative changes in cells of proximal convoluted tulules (X250)

Fig (13) Photomicrograph of portion of renal cortex of curative benazepril treated uraemic rats stained with H&E stain shows tubular cell necrosis and cellular debris in the lumen (x250).