

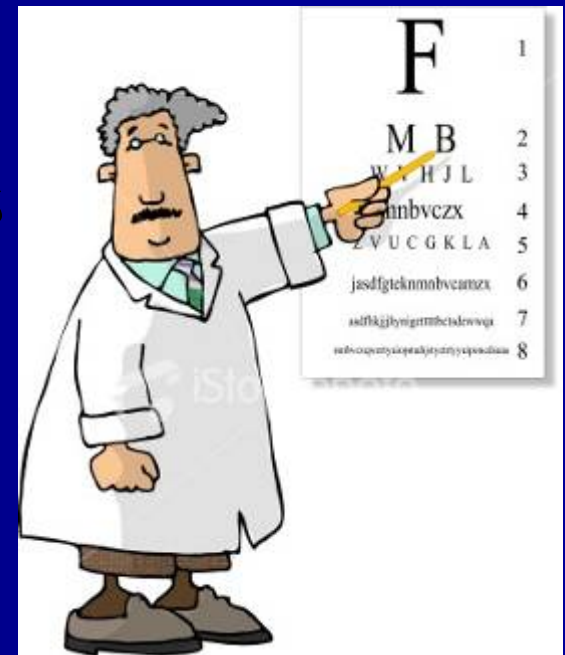
Antioxidant administration with initiation of fever prevents renal scarring and cell apoptosis in the rat model of acute pyelonephritis

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Introduction

- To evaluate the efficacy of antioxidant administration starting by fever detection as one of the first nonspecific presentations of pyelonephritis in reducing renal scars in 2 different models of APN in rat, while comparing effectiveness of Allopurinol and vitamin E in this regard.



Materials and Methods:

- Twenty male rats were allocated into 4 groups. In group 1 to 3, the rats received a direct inoculation of *E-coli* suspension (0.1 ml) into the parenchyma of the right kidney. The rats, in group 4, served as controls and received equal volume of saline.



Materials and Methods:

- ***In group 1 (Abx)*** - daily intraperitoneal (IP) injection of ceftriaxon at a dose of 30 mg/kg once a day started from the third day of bacterial inoculation for 5 days.
- ***In group 2 (Allop.)*** - Allopurinol at a dose of 6.25 mg/kg IP12 was administered as soon as animal's body temperature raised over 37°C and followed administration once a day for five days. When ceftriaxone at a dose of 30 mg/kg once a day started from the third day of bacterial inclusion for 5 days as well.
- ***In group 3 (Vit. E)*** - vitamin E 300 at a dose of 24 IU/kg was given intraperitoneally as soon as animal's body temperature raised over 37°C and followed administration once a day for five days. When ceftriaxone at a dose of 30 mg/kg once a day started from the third day of bacterial inclusion for 5days as well.
- ***In group 4 (Control)*** - Daily intraperitoneal injections of saline at a dose of 10 ml/kg was done for 5 days, started 2 hours following injection of saline to the right kidneys, while no bacterial inoculation and no fever was detected.

Materials and Methods:

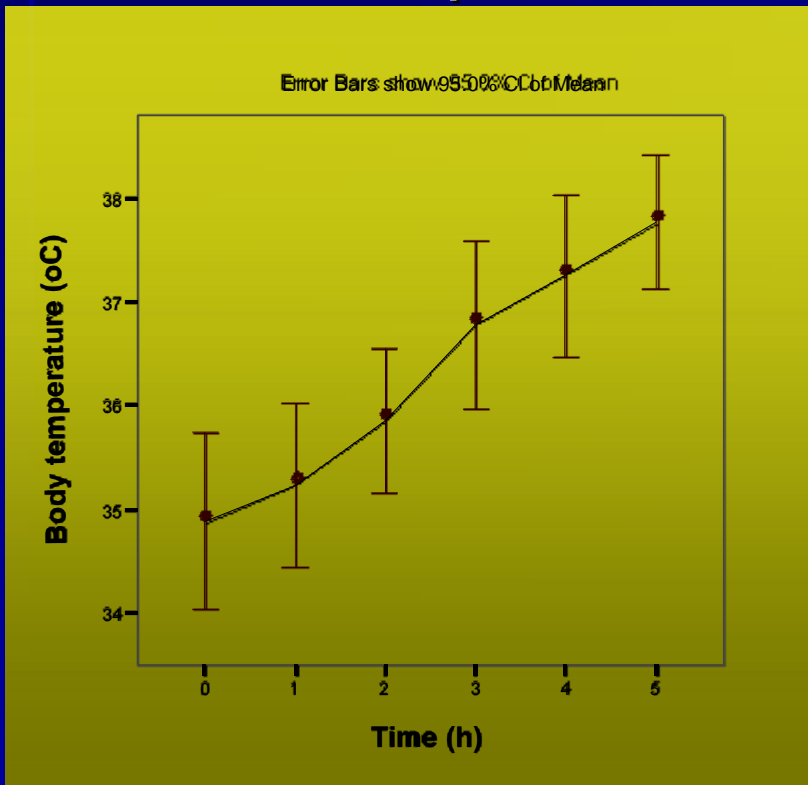
- Six weeks later all rats were scarified and both kidneys examined histopathologically for scarring (Inflammation and fibrosis changes) and cell apoptosis index was determined.



Results:

Body Temperature recordings:

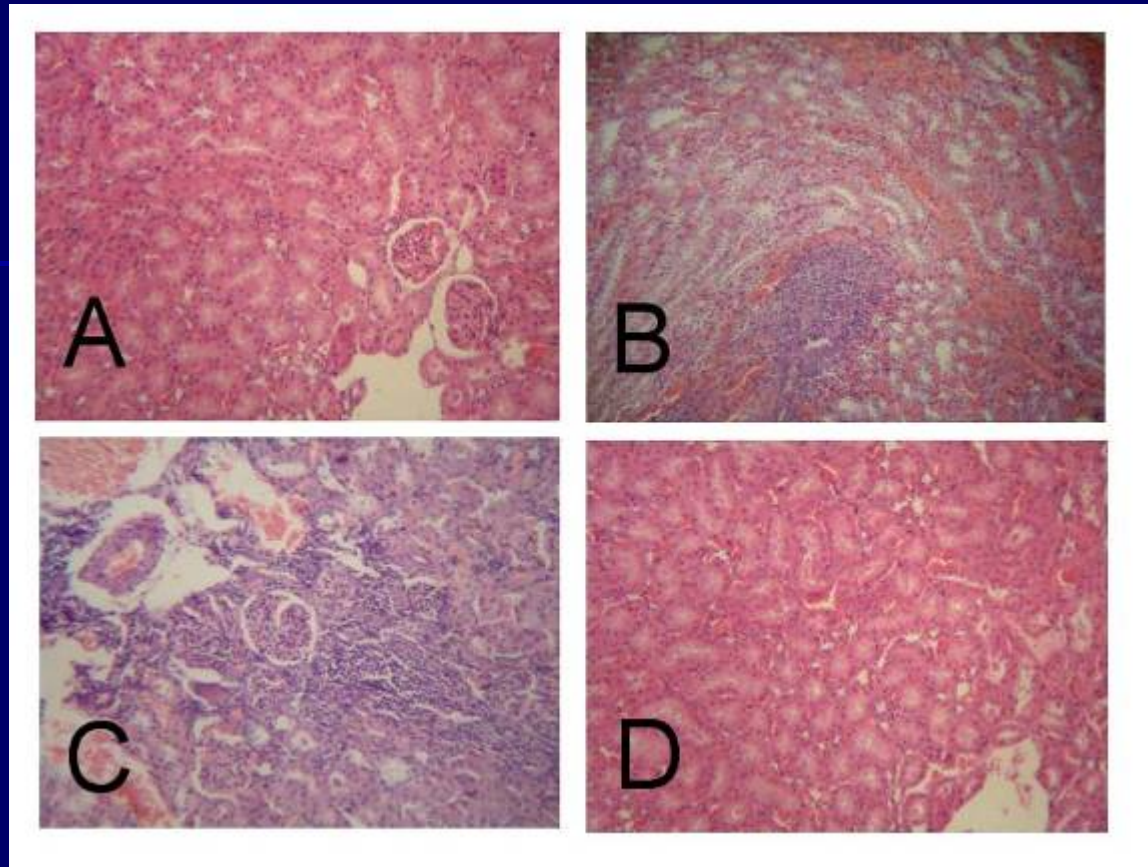
- The fever started 4.6 ± 1.3 hours after inoculation in all rats except one.



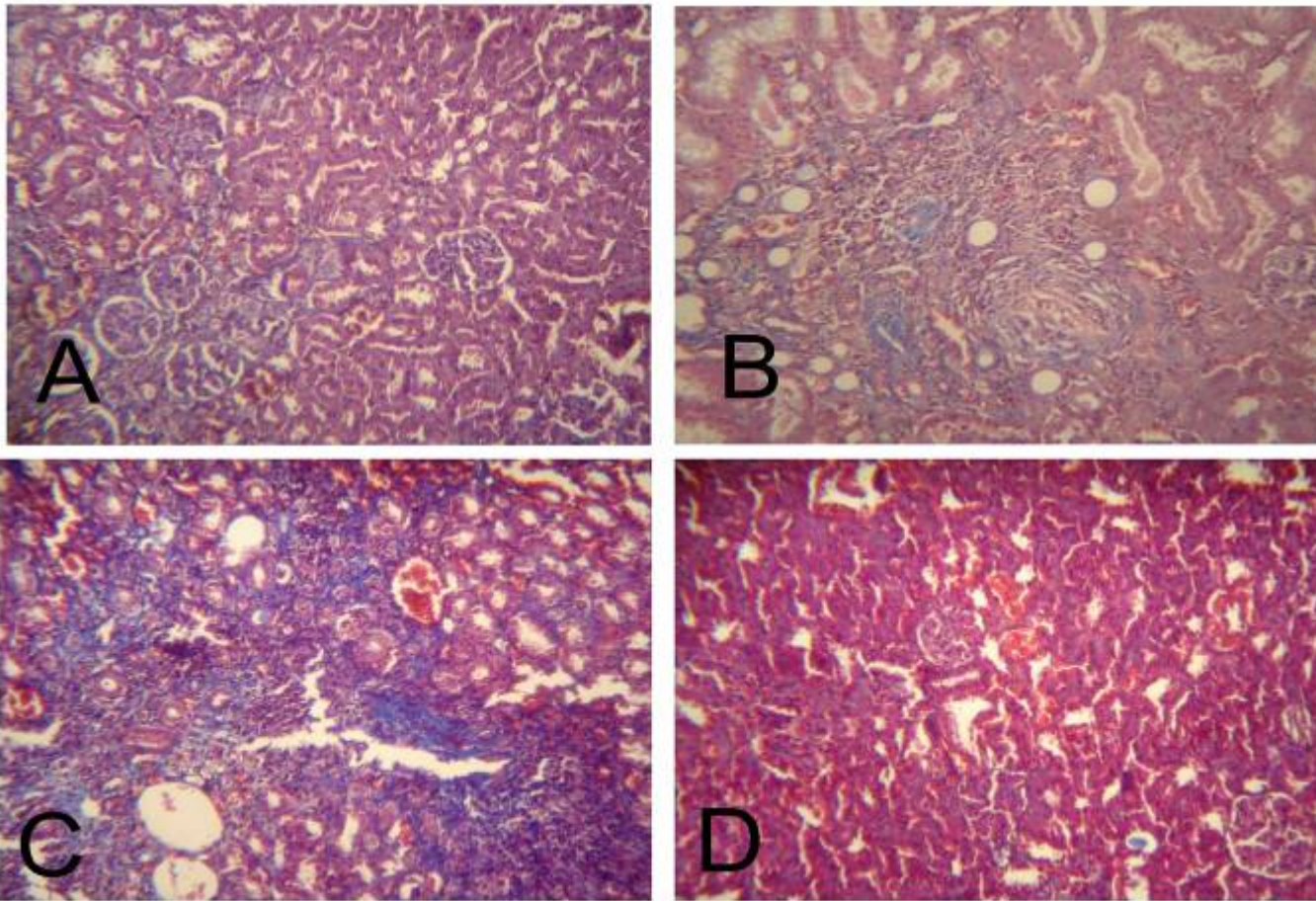
Histopathology Results:

all rats in group 1 showed evidence of renal scar formation. The **inflammation and fibrosis scores** were **higher** in group 1 as compared with Allopurinol and vitamin E treatment groups. Moreover these histopathological changes were significantly **lower** in group 3 compared to group 2 ($p < 0.05$).





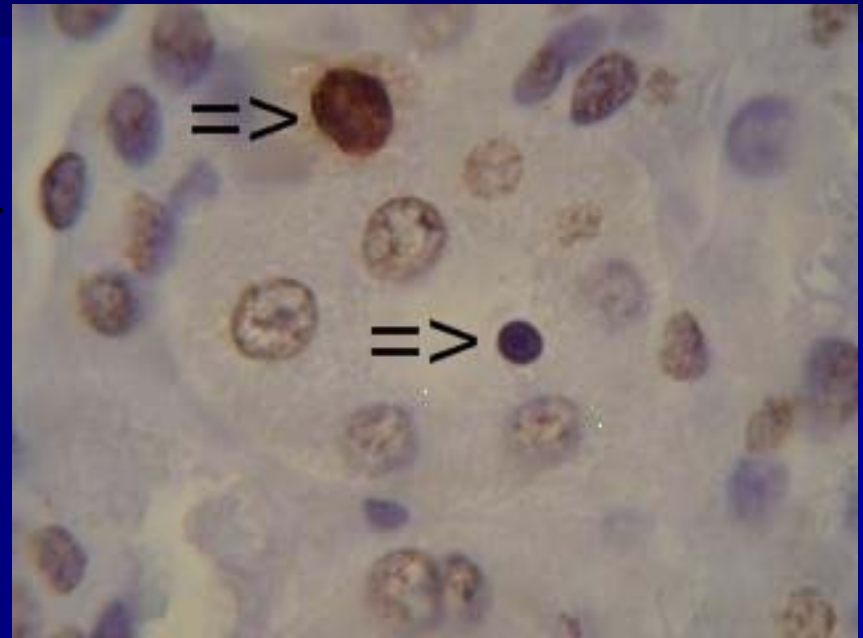
(A) Vitamin E (with initiation of fever) + Antibiotic (started 72 hours post infection) treated pyelonephritis group, shows no parenchymal inflammation or changes; (B) Allopurinol(with initiation of fever) + Antibiotic(started 72 hours post infection) treated pyelonephritis group, shows moderate chronic Inflammation; (C) Antibiotic (started 72 hours post infection) treated pyelonephritis group, shows inflammation and tubular atrophy; (D) Control group, regular renal tissue with glomerulus and tubulus and no inflammation. Microscopic examination staining with HE (100x).



(A) Vitamin E (with initiation of fever) + Antibiotic (started 72 hours post infection) treated pyelonephritis group, shows mild fibrosis; (B) Allopurinol (with initiation of fever) + Antibiotic (started 72 hours post infection) treated pyelonephritis group, shows moderate fibrosis and scarring; (C) Antibiotic (started 72 hours post infection) treated pyelonephritis group, shows severe fibrosis; (D) Control group, regular renal tissue with glomerulus and tubulus and no fibrosis. Microscopic examination staining with Trichrome (100x).

Apoptosis Results:

- The percentage of *apoptotic renal cells* was significantly higher in Abx group (12.09 ± 0.79 %) compared with Vit. E and Allop. groups which had almost same apoptotic index (2.16 ± 0.11 %) ($p < 0.001$)



- All TUNEL-positive cells fulfilling the morphological criteria for apoptosis, including cell shrinkage, nuclear fragmentation, and condensation, were counted. (400x)

Conclusion:

- In summary, our study provided the first clue that initiation of the antioxidant agents following detection of fever can be still effective in reducing the severity of scarring in APN. Moreover, vitamin E seems to be more protective than Allopurinol against renal damage in rats with APN.



References :

- 1. Glauser MP, Meylan P, Bille J. The inflammatory response and tissue damage. *Pediatr Nephrol* 1987; 1:615-19.
- 2. James A, Roberts M, Bernice K, Gary B. Treatment of experimental pyelonephritis in the monkey. *J Urol* 1990; 143:150-54.
- 3. Jodal U. Gentamicin treatments for children with acute pyelonephritis. *Pediatr Nephrol*. 2002; 17:465.
- 4. Pohl HG, Rushton HG, Park J, Chandra R, Majd M. Adjunctive oral corticosteroids reduce renal scarring: The piglet model of reflux and acute experimental pyelonephritis. *J Urol* 1999; 162:815-20.
- 5. Haraoka M, Matsumoto T, Takahashi K, Kubo S, Tanaka M, Kumazawa J. Suppression of renal scarring by prednisolone combined with ciprofloxacin in ascending pyelonephritis in rats. *J Urol* 1994; 151:1078-80.
- 6. Huang A, Palmer LS, Hom D, Anderson AE, Kushner L, Franco I. Ibuprofen combined with antibiotics suppresses renal scarring due to ascending pyelonephritis in rats. *J Urol* 1999; 162: 1396-98.
- 7. Olbing H, Smellie JM, Jodal U, Lax H. New renal scars in children with severe VUR: a 10-year study of randomized treatment. *Pediatr Nephrol*. 2003; 18:1128-31.
- 8. Yagmurlu A, Emin M, Dilek B, Murat E, Dindar H, Ozsan I, et al. Preventive effect of pentoxifylline on renal scarring in rat model of pyelonephritis. *Urology* 2003; 61:1037-41.

References:

- 9. McLorie GA, McKenna PH, Jumper BM, et al. High grade vesicourethral reflux: analysis of observational therapy. *J Urol* 1990; 144:537-45.
- 10. Kavukcu S, Soylu A, Turkmen M, Sarioglu S, Buyukgebiz B, Gure A. The role of vitamin A in preventing renal scarring secondary to pyelonephritis. *BJU Int* 1999; 83:1055-59.
- 11. Bennett RT, Mazzaccaro CRJ, Melman, A, Fanco I. Suppression of renal inflammation with vitamin A and E in ascending pyelonephritis in rats. *J Urol* 1999; 161:1681-84.
- 12. Roberts JA, Laack MB, Fussell EN, Baskin G. Immunology of pyelonephritis VII. Effect of allopurinol. *J Urol* 1986; 136:660-63.
- 13. Singal AK, Bajpai M, Dinda AK. Blockade of Rennin-Angiotensin system blunts the fibrotic response in experimental acute pyelonephritis . *J Indian Assoc Pediatr Surg* 2005; 10:20-24.
- 14. Kaye D. The effect of water diuresis on spread of bacteria through the urinary tract. *J Infect Dis* 1971; 124:297-305.
- 15. Kajbafzadeh AM, Payabvash S, Salmasi AH, Monajemzadeh M, Tavangar SM. Smooth muscle cell apoptosis and defective neural development in congenital ureteropelvic junction obstruction. *J Urol* 2006 176:718.
- 16. Pecile P, Miorin E, Romanello C, Falletti E, Valent F, Giacomuzzi F, et al. Procalcitonin: A marker of severity of acute pyelonephritis among children. *J Pediatr* 2004; 114:249-54.

References :

- 17. Fretzayas A, Moustaki M, Gourgiotis D, Bossios A, Koukoutsakis P, Stavrinadis C. Polymorphonuclear elastase as a diagnostic marker of acute pyelonephritis in children. *J Pediatr* 2000; 105:28-35.
- 18. Chiou Y-Y, Wang S-T, Tang M-J, Lee B-F, Chiu N-T. Renal Fibrosis: Prediction from acute pyelonephritis focus volume measured at 99mTc tmercaptosuccinic acid SPECT. *J Radio* 2001; 221:366-70.
- 19. Lin K-Y, Chiu N-T, Chen M-J, Lai C-H, Huang J-J, Wang Y-T, Chiou Y-Y. Acute pyelonephritis and sequelae of renal scar in pediatric first febrile urinary tract infection. *J Pediatr Nephrol* 2004; 18:362-65.
- 20. Pinson AG, Philbrick JT, Lindbeck GH, Schorling JB. Fever in the clinical diagnosis of acute pyelonephritis. *Americ J Emerg med* 1997; 15:148-51.
- 21. Melis K, Vandevivere J, Hoskens C, Vervaet A, Sand A, Van Acker KJ. Involvement of the renal parenchyma in acute urinary tract infection: the contribution of 99mTc-dimercaptosuccinic acid scan. *Eur J Pediatr* 1992; 151:536-39.
- 22. Rushton HG, Belman AB. Vesicoureteral reflux and renal scarring. In: Holliday M, Barrat M, Anver E, eds. *Pediatric Nephrology*. 3rd ed. Baltimore, MD: Williams & Wilkins; 1994:963-985
- 23. Jakobsson B, Nolstedt L, Svensson L, Soderlundh, Berg U. 99m Technetium dimercaptosuccinic acid scan in the diagnosis of acute pyelonephritis in children: relation to clinical and radiological findings. *Pediatr Nephrol* 1992; 6:328-34.
- 24. Verboven M, Ingels M, Delree M, Piepz A. 99mTc-DMSA scintigraphy in acute urinary tract infection in children. *Pediatr Radiol* 1990; 20: 540-42.