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HEPATO-RENAL ADVERSE EFFECTS OF AMOXICILLLIN AND DOXYCYCLINE IN RATS

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ABSTRACT

Objectives: To evaluate the adverse effects of amoxicillin and doxycycline On liver and kidney functions of rats. **Material and methods:** Twenty-seven male wister albino rats were used and divided randomly into 3 groups' each of 9 rats. Group (1): were served as control and administered 0.5 ml saline orally for 5 consecutive days. Group (2): were served as amoxicillin group and administered 50 mg/kg amoxicillin trihydrate orally for 5 consecutive days. Group (3): were served as doxycycline group and administered 18 mg/kg body weight doxycycline hyclate orally for 5 consecutive days. **Results:** Amoxicillin induced significant increases in serum AST, ALT, direct bilirubin, total protein, creatinine and urea levels. Serum of doxycycline treated rats showed significant increases in serum AST, ALT, total protein and urea levels. Histopathological investigations were further supported the biochemical data of adverse effects on liver and kidney. On conclusion; amoxicillin or doxycycline induced significant damage in liver and kidney, therefore dose should be adjusted especially with patients who suffered from history with liver or renal impairment. Hepatic and renal function tests should be monitored before drug prescription especially for long period. Also, dose and duration of therapy should be adjusted.

KEYWORDS: Amoxicillin, doxycycline, albino rats, histopathology, liver, kidney.

INTRODUCTION

Amoxicillin is a broad spectrum, semi-synthetic, bactericidal penicillin belonged to the β-lactam family (Harvey, 1991).[19] It has been found that it was highly effective against G-positive and G-negative bacteria especially for Helicobacter pylori by inhibiting their cell

wall synthesis (Donowitz and Mandell, 1988; Sahasathin *et al.*, 2007).[11],[33] Excretion of amoxicillin is done predominantly by the kidney by proximal tubules and 10% by glomerular filtration. More than 80% of amoxicillin is recoverable in urine, leading to very high urinary concentrations (Brodie *et al.*, 1990).[8]

Doxycycline is a semisynthetic, broad-spectrum, bacteriostatic tetracycline antibiotic, widely used in veterinary medicine (Fiori *et al.*, 2004).[15] Doxycycline is highly lipid soluble and widely distributed in tissues and fluids. In patients with normal renal function, about 40% of the dose is slowly excreted in the urine. It was reported that doxycycline underwent partial inactivation in the liver. Also, accumulation of doxycycline in patients with renal failure has been reported (Martindale, 2009).[26]

Both antibiotics are necessary drugs, widely used and there were insufficient data about their adverse effects On liver and kidney, therefore the purpose of the present study was to study the hepato-renal adverse effects of amoxicillin and doxycycline in rats.

MATERIALS AND METHODS

Materials

Amoxicillin trihydrate obtained from SINOPHARM WEIQIDA PHARMACEUTICAL CO., Ltd, CHINA.

Doxycycline hyclate obtained from ATCO PHARMA FOR PHARMACEUTICAL INDUSTRIES, EGYPT.

Laboratory animal

Twenty-seven male wister albino rats (200-250g) were obtained from department of laboratory animals, Faculty of Veterinary Medicine, Benha University, Egypt. Rats were housed in stainless steel wire mesh cages with bedding of ground wood chips. Rats were kept at a constant environmental and nutritional conditions throughout the period of experiment. The animals were left for 14 days for acclimatization before the beginning of the experiment.

METHODS

The rats were randomly divided into three groups, each of 9 rats. Group (1): Nine rats were served as control and were administered saline only (0.5 ml orally for 5 consecutive days). Group (2): Nine rats were served as amoxicillin group and were administered amoxicillin trihydrate. (50 mg/kg b. wt. orally for 5 consecutive days). Group (3): Nine rats were served

as doxycycline group and were administered doxycycline hyclate (18 mg/kg b.wt. orally for 5 consecutive days).

Blood samples were taken after first, seventh and fourteenth days post-treatment in all groups (after the end of administration of amoxicillin and doxycycline). Blood samples were collected without anticoagulant. Serum was obtained by high speed centrifugation of blood at 1000 g for 15 minutes and kept at 4 ^{o}C in a refrigerator, until analysis. Serum samples were used for quantitative determination of serum bilirubin, serum aspartate aminotransferase activity, serum alanine aminotransferase activity, serum gamma glutamyl transferase, serum total protein level, serum albumin level, serum creatinine, serum urea and serum uric acid.

The experimental rats were sacrificed at first, seventh and fourteenth day after drug administration; tissue samples from liver and kidney were collected for histopathological examination.

Histopathological examination of liver and kidney

The preparation of liver and kidney samples and procedures of staining were carried out according to Banchroft *et al.* (1996).[2]

Statistical analysis

The data were calculated as mean ± standard error. All statistical analysis was carried out using Students paired *t*-test to express the differences between groups according to Berly and Lingren (1990)[4], comparison of the mean values was performed, and the differences were considered statistically significant when $P < 0.05$.

RESULTS

Effect on biochemical parameters

Amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) induced significant increases in serum AST, ALT, direct bilirubin, total protein, creatinine and urea levels. Doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) induced significant increases in serum AST, ALT, total protein and urea levels. These significant changes are recorded in figures.[1- 6]

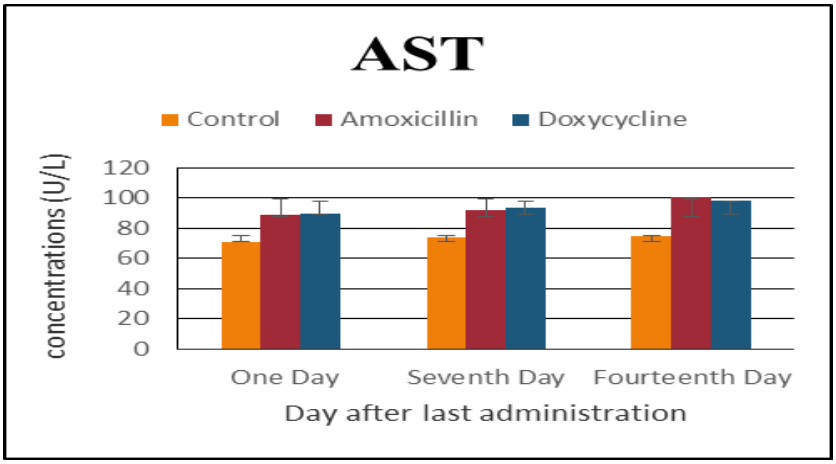

Figure 1: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum aspartate aminotransferase level (U/L) in rats (n=4).

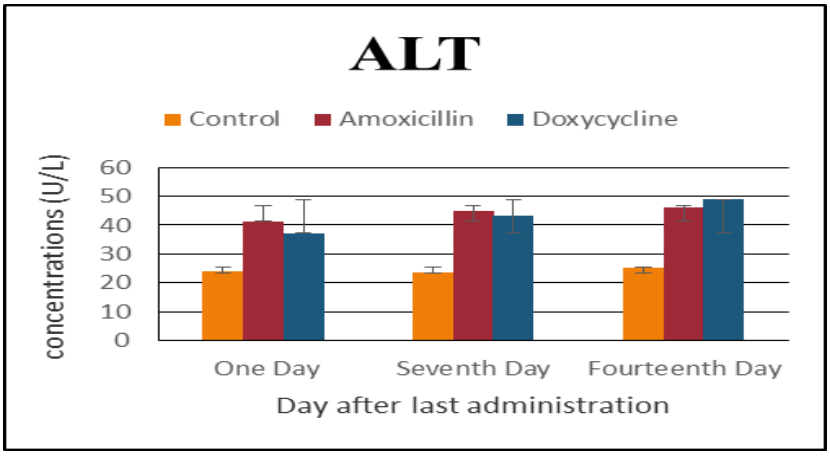

Figure 2: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum alanine aminotransferase level (U/L) in rats (n=4).

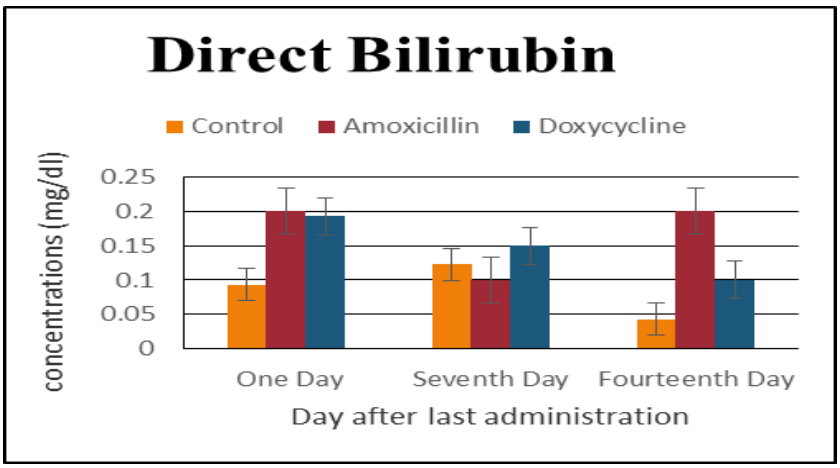

Figure 3: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum direct bilirubin level (mg/dl) in rats (n=4).

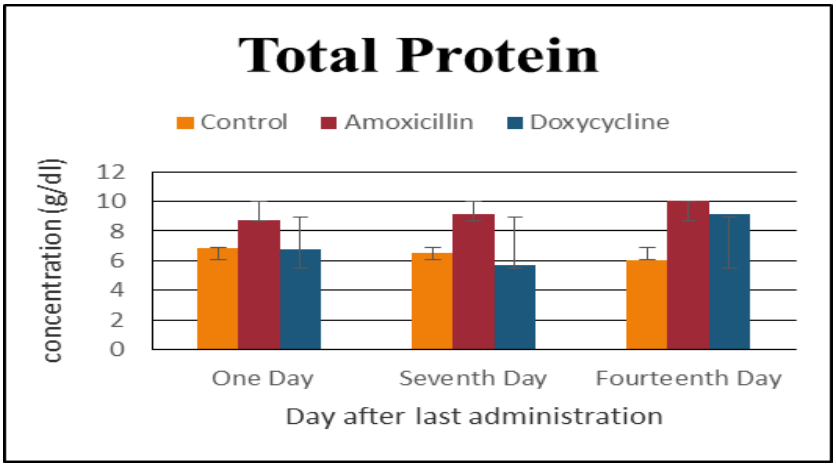

Figure 4: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum total protein level (g/dl) in rats (n=4).

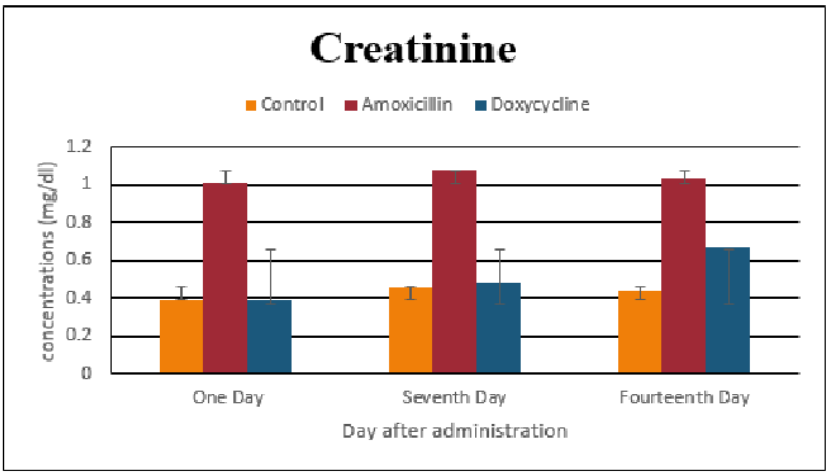

Figure 5: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum creatinine level (mg/dl) in rats (n=4).

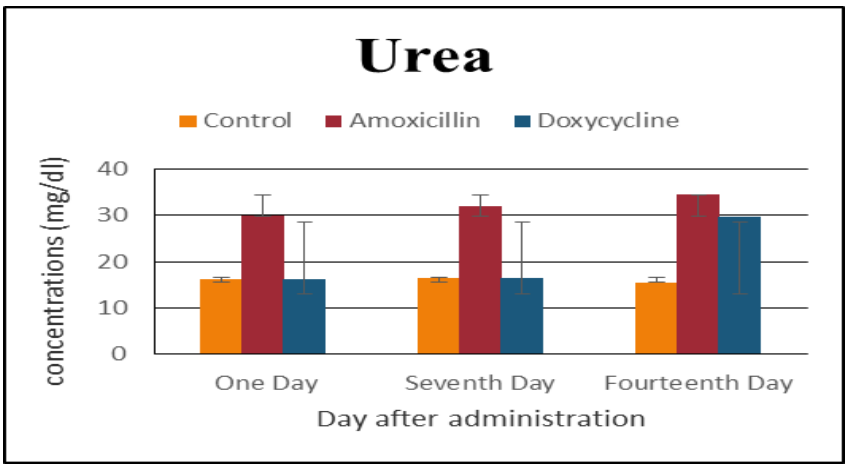

Figure 6: Effect of amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) and doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) on serum urea level (mg/dl) in rats (n=4).

Histopathological findings

Amoxicillin administration (50 mg/kg b.wt. orally daily for 5 consecutive days) induced dilatation of central vein with degeneration in surrounding adjacent hepatocytes in liver [Figure 7]. Vacuolization in lining endothelium of the glomerular tuft with degeneration in lining epithelium of some individual tubules, inflammatory cells infiltration and focal extravasation of red blood cells in between renal tubules seen in kidney [Figure 8].

Doxycycline administration (18 mg/kg b.wt. orally daily for 5 consecutive days) induced dilatation of central vein associated with degeneration in the surrounding adjacent hepatocytes [Figure 9a] with inflammatory cells infiltration in the portal area [Figure 9b]. In kidney, vacuolization of the endothelial cells lining the tuft of the glomeruli with focal extravasation of red blood cells in between renal tubules was found [Figure 10].

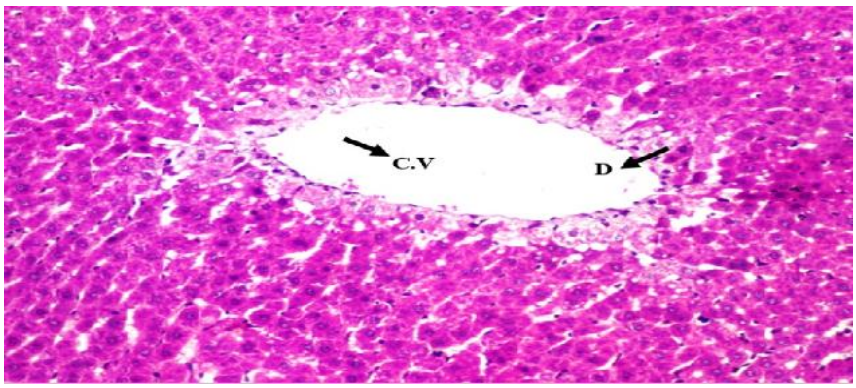

Figure 7: Liver of treated rat with amoxicillin 50 mg/kg b.wt. orally daily for 5 consecutive days showing dilatation of central vein (c.v) with degeneration in surrounding adjacent hepatocytes (D). (H&E, x40).

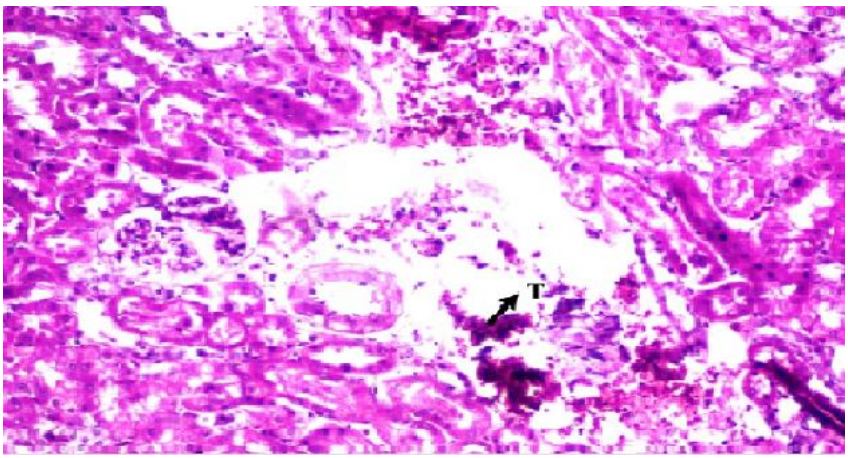

Figure 8: Kidney of treated rat with amoxicillin 50 mg/kg b.wt. orally daily for 5 consecutive days showing vacuolization in lining endothelium of glomerular tuft with degeneration in lining epithelium of some individual tubules, inflammatory cells infiltration and focal extravasation of red blood cells in between renal tubules (T). (H&E, x40).

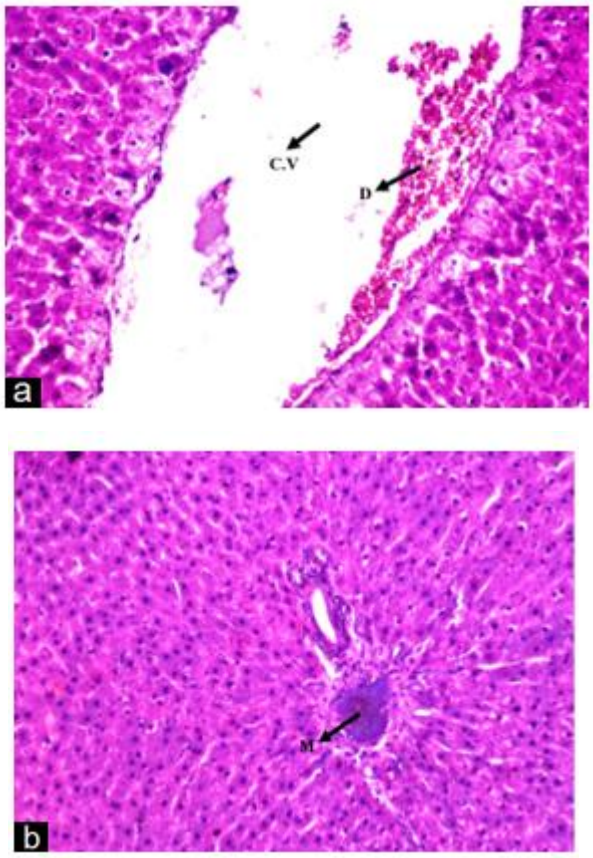

Figure 9: Liver of treated rat with doxycycline 18 mg/kg b.wt. orally daily for 5 consecutive days showing (a) dilatation of central vein (c.v) associated with degeneration (D) in the surrounding adjacent hepatocytes. (H&E, x40). (b) inflammatory cells infiltration in the portal area (M). (H&E, x40).

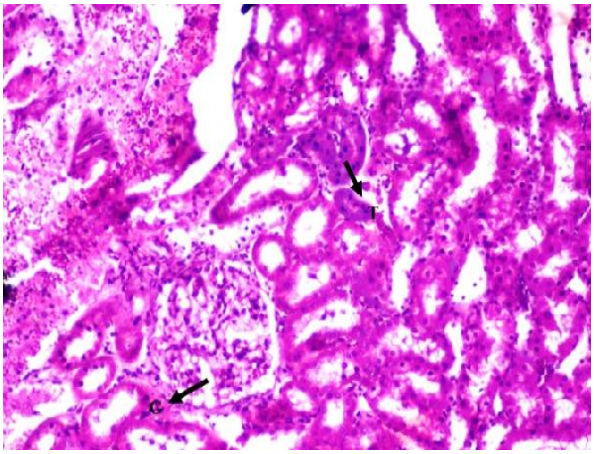

Figure 10: Kidney of treated rat with doxycycline 18 mg/kg b.wt. orally daily for 5 consecutive days showing vacuolization of the endothelial cells lining the tuft of the Glomeruli (G) with focal extravasation of red blood cells in between renal tubules (T). (H&E, x40).

DISCUSSION

In the present study, it has been observed that amoxycillin or doxycycline induced renal and liver damage in rats. This was evident from the renal and liver function tests suggesting impairment of renal and liver function.

Amoxicillin treated rats (50 mg/kg body weight orally for 5 consecutive days) showed significant increases in serum direct bilirubin, serum aspartate aminotransferase, serum alanine aminotransferase and total protein levels, may be due to the release of enzymes from hepatic tissues due to hepatic damage. The obtained results were in accordance with the results of histopathological study as dilatation of central vein and degeneration of hepatocytes. which agreed with Larrey et al., (1992),[21] Farrell, (1995),[14] Hautekeete et al., (1995),[18] Renner, (1995),[31]Rodriguez et al., (1996),[32] Tredger and Sherwood, (1997),[38] Limauro et al., (1999),[22] Macfarlane *et al.*, (2000),[26] Amr and Alaa, (2005),[1] Kikkawa *et al.*, (2006),[20] and Olayinka and Olukowade, (2010).[28]

The significant increases in serum creatinine and serum urea levels indicated impaired renal function in filtration of waste products attributed to renal insufficiency which may be due to vacuolization of glomerular tuft and degeneration of renal tubules as observed in histopathological study. The obtained results were coordinated with the results obtained by Edward *et al.*, (1974),[12] Geller *et al*., (1986),[16] Cameron and Greger, (1998),[10] Orth and Ritz, (1998),[30] Olayinka and Olukowade, (2010),[28] and Elmajdoub *et al.*, (2014).[13]

Doxycycline treated rats (18 mg/kg body weight orally for 5 consecutive days) showed significant increases in serum aspartate aminotransferase, serum alanine aminotransferase and total protein levels. The elevation in serum aspartate aminotransferase and serum alanine aminotransferase may be due to the release of the enzymes from hepatic tissues due to hepatic damage as in histopathological studies where degeneration of hepatocytes and inflammatory cells infiltration in portal area were observed. The obtained results were in accordance with the results of Böcker *et al.*, (1982),[7] Hopf *et al.*, (1985),[17] Bjornsson et al., (1997),[5] Macfarlane *et al.*, (2000),[23] Machado et al., (2003),[24] Kikkawa et al. (2006),[20] Schulz *et al.*, (2011),[34] and Shabana *et al.*, (2012).[35]

The significant increase in urea level may be due to impaired renal function where swelling and vacuolization of endothelial cell lining of glomeruli were observed in histopathological study. The present results agreed with those obtained by Shils, (1962,1963),[36],[37]

Mavromatis, (1965),[25] Orr *et al.*, (1978),[29] Hopf *et al.*, (1985),[17] Bihorac *et al.*, (1999),[3] Chambers, (2001),[9] Muthukumar *et al.*, (2002),[27] Machado *et al*., (2003),[24] Zallen, (2009),[39] and Shabana *et al.*, (2012).[35]

CONCLUSION

Regarding the wide uses of amoxicillin or doxycycline drugs as a safe choice in most cases. However, both drugs can cause severe damage in liver and kidney especially with patients who suffered from liver or renal impairment. Hepatic and renal function tests should be monitored before drug prescription especially for long period. Also, dose and duration of therapy should be adjusted.

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