Research Article

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Protective Effects of Vitamin D3 on Male Rat's Hepatic Cell Injured by Diclofenac, A Histopathological Study

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Abstract

This experiment aimed to study the protective effects of vitamin D3 on hepatic cells injured by Diclofenac. Twenty-one male rats, 8 to 12 weeks of age and in weight from 180 to 220 grams, were housed in a standard, pathogen-free environment in the "College of Veterinary Medicine, University of Fallujah," given unrestricted access to standard rat water and food. Then the rats were divided into 3 groups (each group containing 7 rats): G1 control group (IM injection of distill water for 7 days), G2 Diclofenac only (7-day course of a 10 mg/kg IM injection of diclofenac), G3 received intramuscular injections of VD3 three times per week at a dose of 1000 IU/Kg, then a 7-day course of a 10 mg/kg IM injection of diclofenac. Liver histological sections of the normal control (G1) showed no histopathological changes, while the histopathological sections of the liver of G2 exhibit hydropic degeneration, blood vessels congestion and inflammatory cell infiltration, Liver sections of G3 presented blood vessels congestion and mild inflammatory cell infiltration. This study concluded that VD3 can decrease the liver damage caused by diclofenac

Keywards: Diclofenac, liver, histopathological changes, Vit D3.

التأثيرات الوقائية لفيتامين دي 3 على الخلايا الكبدية لدى ذكور الجرذان التي تأثرت بالديكلوفيناك ، دراسة نسيجية مرضية

الخلاصة:

هدفت هذه التجربة إلى دراسة التأثيرات الوقائية لفيتامين D3 على الخلايا الكبدية المتأثرة بالديكلوفيناك. تم استخدام واحد وعشرون من ذكور الجرذان لهذه التجربة تتراوح أعمارهم من 8 إلى 12 أسبوعًا ويزن من 180 إلى 220 جرامًا، تم إيواؤهم في بيئة قياسية خالية من مسببات الأمراض في كلية الطب البيطري / جامعة الفلوجة ، مع منحهم كميات غير محددة من الماء والطعام. ثم قسمت الجرذان إلى 3 مجموعات (كل مجموعة تحتوي على 7 جرذان): مجموعة التحكم G1 (حقن عضلي من الماء المقطر لمدة 7 أيام) ، المجموعة الثانية G2 ديكلوفيناك فقط (دورة لمدة 7 أيام لحقن 10 مجم / كجم عضلي من ديكلوفيناك)، اما المجموعة الثالثة G3 (ديكلوفيناك مع فيتامين دي 3) فتم حقنها عضليا من فيتامين دي 3 ثلاث مرات في الأسبوع بجرعة 1000 وحدة دولية / كجم ، ثم دورة لمدة 7 أيام من 10 مجم / كجم من ديكلوفيناك بواسطة الحقن العضلي ايضا. لغرض الدراسة النسجية والمرضية,. لم تظهر المقاطع النسيجية للكبد والمأخوذة من مجموعة السيطرة (G1) أي تغيرات نسيجية مرضية, بينما تظهر المقاطع النسيجية للكبد والمأخوذة من المجموعة الثانية G2 تنكسًا مائيًا واحتقانًا في الأوعية الدموية وتسللًا للخلايا الالتهابية, اما فيما يخص المقاطع النسيجية للكبد المأخوذة من المجموعة الثالثة G3فقد أظهرت احتقان الأوعية الدموية وتسلل الخلايا الالتهابية الخفيفة. استنتجت هذه الدراسة أن فيتامين دي 3 يمكن أن يقلل من تلف الكبد الناجم عن استخدام ديكلوفيناك.

Introduction:

Non-steroidal anti-inflammatory drugs (NSAIDs) are often used to alleviate pain as well as inflammation, mild side effects are seen at or produced by standard therapeutic dosages, however, these medications have a high toxicity level when used in large quantities. Many NSAIDs work by blocking cyclooxygenase enzyme 2 (COX2) (1), which in turn alters prostaglandins synthesis (2) and puts kidney and liver cells at risk of damage. (3).

Diclofenac (DF) is NSAID that contains phenylacetic acid and has the ability to reduce inflammation, relieve pain, reduce fever. (4,5). While DF has useful therapeutic applications, it also comes with a number of potentially life-threatening risks. Toxic effects on the digestive system, as well as harm to the cardiovascular system, liver, lungs and kidneys, making it a non-threshold multitargeted medication (6, 7, 8).

Although there is proof that Diclofenac (DF) can harm mitochondria by upsetting immune-mediated defenses, producing reactive oxygen metabolites, and suppressing the activity of "enzymatic and nonenzymatic antioxidants in kidney and liver tissues, the precise mechanism of DF-caused kidney as well as liver toxicity is still unclear (9, 10, 11)."

Vitamin D3 and vitamin D2 belong to a class of seco-steroid compounds that are referred to as vitamin D. (12). It is considered an organic compound found in food and is required for health and the musculoskeletal system in trace amounts(13)

Vitamin D3 (also known as valproic acid, or VD3) is a steroid hormone that is typically obtained either through exposure to ultraviolet sunlight on the skin or through the consumption of food high in VD3. VD3 undergoes two hydroxylations in the liver and one in the kidneys to become physiologically active by the enzymes "vitamin D 25-hydroxylase (Cyp2r1) and vitamin D 1 hydroxylase (Cyp27b1)". This causes calcitriol to take on its active form (14).

This investigation aimed to study the Protective effects of vitamin D3 on hepatic cell injured by Diclofenac.

Materials and methods:

Twenty-one male rats, ranging from 8 to 12 weeks in age and weightiness from 180 to 220 grams, were housed in a standard, pathogen-free environment in the "College of Veterinary Medicine, University of Fallujah," and given unrestricted access to standard rat food as well as water. These were divided into three groups (each group contain 7 rats):

G1 control group (IM injection of distill water for 7 days)

G2 Diclofenac only (7-day course of a 10 mg/kg IM injection of diclofenac.) (Acino company, Swiss).

G3 The VD3 group received intramuscular injections of VD3 three times per week at a dose

of 1000 IU/Kg (15), then a 7-day course of a 10 mg/kg IM injection of diclofenac. (DAWAAI, Pakistan)

For histological analysis, the liver sections of each experimental group were fixed in 10% formol-saline and prepared by Routine histological techniques. Slides were stained by Hematoxylin- eosin stain (16).

Results and discussions:

Liver sections of the control group showed no clear pathological lesions, Examination of liver sections in the control group (G1) showed no clear pathological lesions, normal polygonal hepatocytes with mixed euchromatic or heterochromatic nuclei and limited number of binucleated cells. Sinusoids architectures were normal (Fig.1), while histopathological sections of liver of G2 exhibit disappearance of the limits of some hepatocytes, hydropic degeneration, blood vessels congestion and inflammatory cell infiltration (Fig.2), histopathological sections of liver of G3 showed milder disturbance of liver architecture than G2 presented by blood vessels congestion and mild inflammatory cell infiltration (Fig.3).

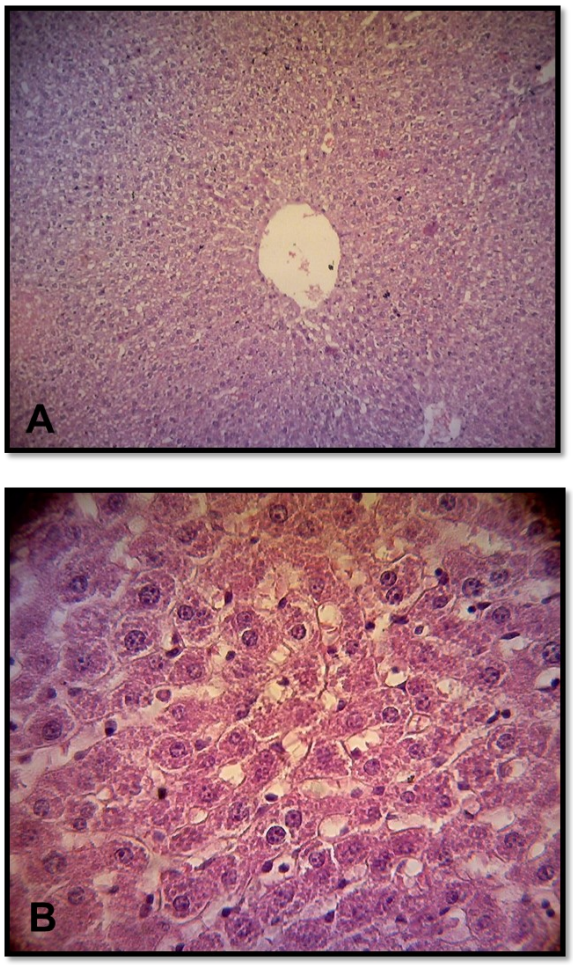

Figure 1. Histological image of liver sections from control group (G1) Showed no clear pathological changes, normal architecture of hepatocytes and sinusoids, no blood vessels congestion and no inflammatory cell infiltration H&E AX10, BX40.

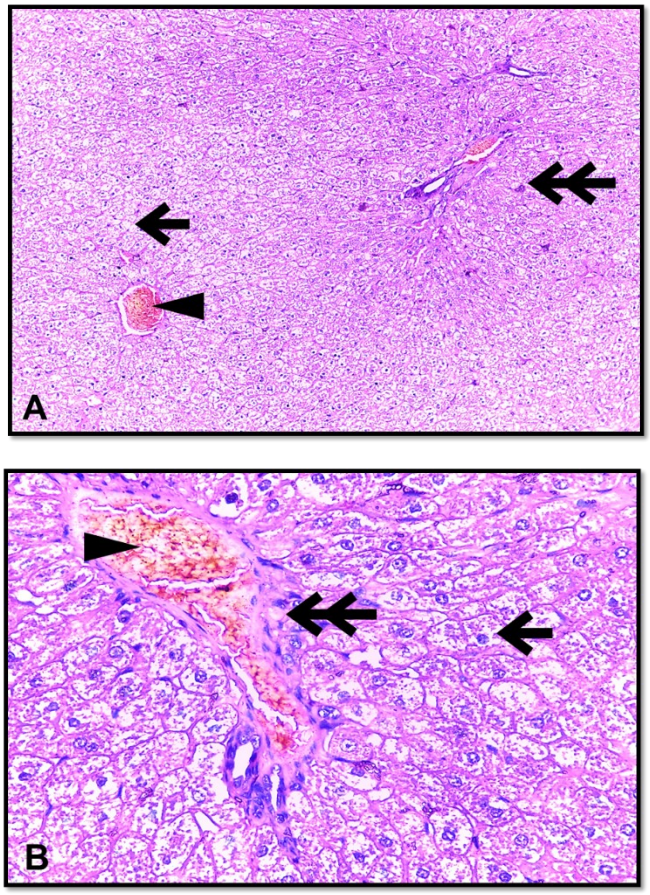

Figure 2. Histopathological image of liver from treated with diclofenac (G2) showed disappearance of the limits of some hepatocytes, hydropic degeneration (arrow), blood vessels congestion(arrowhead) and inflammatory cell infiltration (double arrow). H&E AX10, BX40.

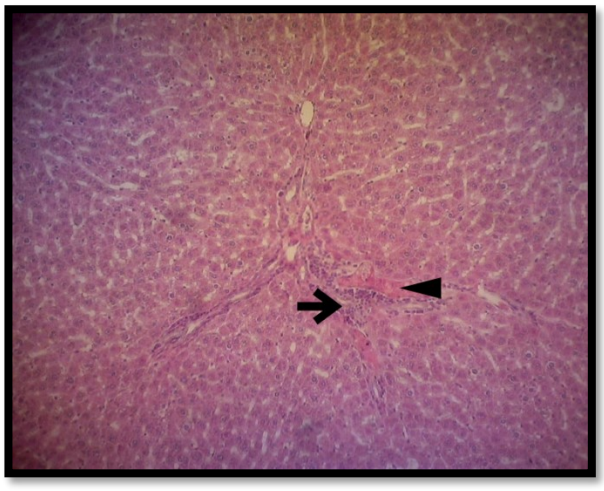

Figure 3. Histopathological image of liver from treated with diclofenac with vit D3 (G3) showed blood vessels congestion (arrowhead) and mild inflammatory cell infiltration (arrow). H&E X10.

The present study was in agreement with results of (17) who found that "examining the liver tissues of rats given diclofenac sodium at 100 and 150 mg/kg for 14 days revealed severe wounds that worsened with increasing drug dose, including widespread degeneration and vacuolation, and peri-acinar rot with mild to severe invasion of gateway zones with mononuclear cells". Also, the results was compatible with those of (18) who found that liver manifest cellular deterioration, necrosis, vascular dilatation, lobular congestion, portal enlargement, and inflammatory cell infiltration around necrotic hepatocytes and the portal region after DF treatment.

It had been reported that, Vit. D3 may help reverse liver damage after long-term NSAIDs usage because to its protective effects on hepatocytes (19, 20), these was in agreement with the results of current study. Additionally, present study results were the same of research done to study the effects of NSAIDs on the kidney, liver, and testicles and the protective role of D3, the results Proved the protective effects of VD3 in cases of toxicity by NSAIDs (21). The ability of vitamin D3 (cholecalciferol) to keep hepatocytes functioning normally may help to repair liver damage brought on by chronic NSAIDs treatment. 1,25-dihydroxyvitamin D3 (1,25(OH)2D3, calcitriol), the hormone-active metabolite of vitamin D3, and the nuclear vitamin D3 receptor (VDR) work on target cells to control several essential biological activities. Bone remodeling and calcium and phosphorus

metabolism modulation are not the only things this hormone does (22).Also, it has been reported that when compared to the prednisolone group, the vitamin D3 group had much less NO production in hepatocytes (23) which prove the decreasing damage on hepatic tissues when treated by D3 in current study

Conclusion:

The present study concluded that VD3 had a role to decrease the liver damage induced by diclofenac.

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Confect of Interest

There is no conflict of interest

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