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# The protective role of vitamin c against the hepatotoxic and nephrotoxic effect of meloxicam in male mice

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***Abstract:*** *An ameliorating effect of vitamin C on the toxic effect of meloxicam, a new non-steroidal anti-inflammatory drug was studied by evaluating biochemical parameters, histopathological changes in liver and kidney of mice. A total of forty male mice were divided in four experimental groups each comprising of ten mice Meloxicam toxicity was induced by oral feeding of meloxicam at 0.4 mg/kg in G3 and G4 respectively for 28 days. Group G4 was fed with 0.4mg/kg body weight of meloxicam along with 350 mg/kg body weight of vitamin C. Group G1 serve as control while group G2 was fed only vitamin C at 350 mg/kg body weight. Tissue alkaline phosphatase(ALP), alanine transaminase(ALT), aspartate transaminase (AST) were found elevated in meloxicam treated groups and indicated hepatotoxic activity of meloxicam. Vitamin C was succeeded in reducing hepatotoxic activity of meloxicam in group G4 receiving meloxicam along with vitamin c compared with meloxicam treated group. In meloxicam toxicity, elevated malondialdehyde (MDA) value was noticed in liver homogenate. Furthermore, necrosis with infiltration with inflammatory cells was evident in liver and kidney. The vitamin c could protect the liver and kidney against the toxic effect of meloxicam.*
***Keywords:*** *Meloxicam, Vitamin c, Liver, Kidney, Mice*

## I. Introduction

Mobic or meloxicam, a non-steroidal anti-inflammatory drug (NSAID) is widely used in humans, cattle, buffalo, goats and dogs. Meloxicam is a COX-2 (cyclo-oxygenase) inhibitor at its lowest therapeutic dose and is an anti-inflammatory by inhibiting prostanoid synthesis in inflammatory cells[1]. Meloxicam inhibits COX-2 about 12 times more selectively than COX-1[2]. Most NSAIDs are metabolized in the liver by oxidation and converted to inactive metabolites, which are then typically excreted in the urine while some drugs are partially excreted in bile. Meloxicam, as NSAID, either alone or with antimicrobial drugs, is indicated for use in ruminants for the treatment of pneumonia, pleuritis, laminitis, myositis, sprain, mastitis, prolapse of uterus, premature labour etc. The principle side effect of meloxicam is gastrointestinal irritation (vomiting, diarrhea and ulceration), but important side effects include liver and kidney toxicity were recorded [3, 4, 5 , 6]. The action of action of meloxicam toxicity is through oxidation-antioxidation mechanism in which it is considered oxidative stress inducer [7,8] but others found it acts as antioxidant [9,10].

Besides exerting antioxidant influence directly [11], VC can promote the repair of oxidative DNA damage from the DNA and/or nucleotide pool, through upregulation of repair enzymes [12]. Therefore we aimed to use this vitamin to protect the liver and kidney against the toxic effect of meloxicam.

## II. Material And Methods

Meloxicam was purchased from Boehringer Ingelheim Gmb& CoKG. Vitamin C was purchased from Sigma, USA. All the biochemical kits and chemicals used in the study were of analytical grade. Forty male Swiss mice were divided into four groups of 10 animals each. All the animals were maintained in polypropylene cages, (47 × 34 × 18 cm). The maximum number of animals in each cage was six. Dried rice husk was used as a bedding material. Bedding material was changed every alternate day. All the animals were kept under standard managemental conditions as per the norms of Committee for the purpose of control and supervision on experiments on animals (CPCSEA), and were maintained under a controlled environment with temperature at 23± 2°C, relative humidity at 55 ± 5%, and a 12-hours/12 hours light/dark cycle throughout the experiment. The animals were fed on a standard pellet diet. The pellet diet consisted of 22.02% crude protein, 4.25% crude oil, 3.02% crude fiber, 7.5% ash and 1.38% sand silica. They were given ad-lib feed and whole some drinking water throughout the experiment. The necessary Institute Animal Ethical committee approval was obtained.

The mice were acclimatized for 15 days to the environment, before the start of the experiment. A total of forty male mice, were divided in to four groups (G1, G2, G3 and G4) with 10 mice in each group. Group 'G1' served as control and was treated with vehicle, 0.5 distilled water. Group 'G2' received 350mg/kg vitamin c, 'G3', which received meloxicam at 0.4 mg/kg body weight and 'G4' received meloxicam at 0.4mg/kg body weight plus 350mg /kg body weight vitamin C.

The mice from all experimental groups were sacrificed on 24th day of experiment. Prior to sacrifice the

mice were fasted for 12 hours. Animals were sacrificed by chloroform anesthetization at the end of experiment.. The representative tissues – liver and kidney were fixed in 10% Bouins fluid immediately on removal. They were subjected for histopathological examination following standard procedure[13]. Liver pieces were collected in ice cold containers for estimation of tissue ALT,AST,ALP using analytical kits supplied by BIOLABO SA 02160MAIZY –FRANCE )and malondialdehyde (MDA) level by the method of Kartha and Krishnamurthy, (1977)[14].

The data generated was analyzed with one way analysis of variance (ANOVA). The level of significance was set at $P < 0.01$.

## III. Results And Discussion

As shown in Table(1), meloxicam caused significant elevation in the level of malondialdehyde (MDA) in mice liver homogenate compared to control and vitamin c alone, while a significant decrease has been recorded in the meloxicam treated group after treating with vitamin c in comparison to meloxicam alone. This indicates that meloxicam induce oxidative stress since MDA, which is the last product in lipid peroxidation process, is considered as oxidative stress marker[15]. Similar elevation in MDA level in response to meloxicam treatment was recorded in previous studies [5,16]. With respect to liver enzymes, meloxicam has found to cause significant elevation in the level of ALT, AST and ALP in the liver homogenate, while a significant decrease 2 was detected in the meloxicam plus vitamin c group in comparison to meloxicam alone. These results came in agreement with other study conducted by Al-Rekabi et al.,2009)[4]. From such changes in liver enzymes, we can suggest a hepatotoxic effect for meloxicam[5].

Histologically, control group showed normal liver structure(Fig1), while meloxicam caused degeneration of hepatocytes(mainly necrosis), dilatation in blood sinusoid lumen and appearing of inflammatory leucocytes aggregations (Fig 2). Vitamin c alone didn't show such obvious histological changes. After treating the meloxicam treated mice with vitamin c, a clear attenuation has been obtained with approximately normal structure of liver structure, although few degenerated cells were still seen(Fig 3). Such attenuative effect of vitamin c against meloxicam toxicity is explained by the oxidation –antioxidation mechanism since vitamin c is a strong antioxidant agent [17, 18].

Several histological alteration were detected in the kidney of mice treated with meloxicam such as congestion of blood vessels with blood cells, appearance of inflammatory cells aggregates and degeneration of epithelial cells lining the kidney tubules in comparison with normal structure of kidney in control group(Fig 4 and 5). Similar nephrotoxic effect of meloxicam was previously detected [6]. On the other hand, vitamin c was succeeded in protecting the kidney from these changes after treating the meloxicam treated mice with the vitamin(Fig 5).

## IV. Conclusion

From the present investigation it can concluded that meloxicam caused hepatotoxic and nephrotoxic effect from biochemical and histological points of view and vitamin c may exert a protective effect against these effects.

## References

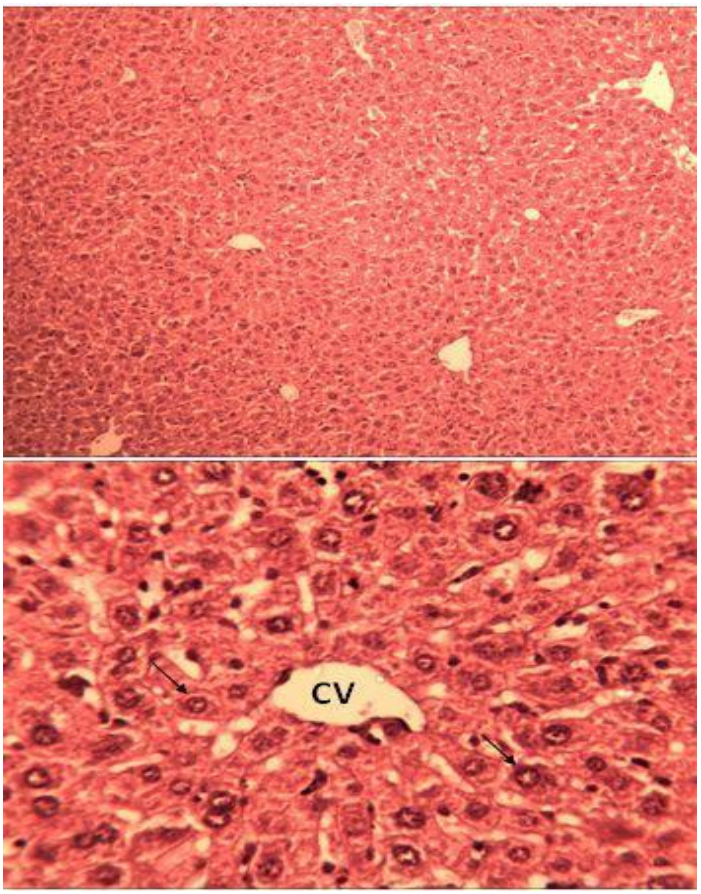
[1] R. Fleischmann, I. Iqbal, G. Slobodin, Meloxicam, Expert Opinion Pharmacotherapy Journal,3,2002, 1501–12.
[2] K. Ogino, Y .Harada, K. Hatanaka, The 69th Annu. Meet Nagasaki, Japan, Inhibitory effect of Meloxicam on COX-2. Japanese Pharmacological Society. 1996.
[3] JR. Vane, RM. Botting, Mechanism of action of aspirin-like drugs. Semin Arthritis Rheumtology, 26(6 Suppl 1) ,1997, 2–10
[4] FM. Al-Rekabi, DA. Abbas and NR. Hadi, Effects of subchronic exposure to meloxicam on some hematological, biochemical and liver histopathological parameters in rats, Iraqi Journal of Veterinary Sciences, 23, Supplement II, 2009 (249-254)
[5] R. Mahaprabhu, A G. Bhandarkar, BL. Jangir, S P. Rahangadale, and N V. Kurkure, Ameliorative effect of *Ocimum Sanctum* on meloxicam induced toxicity in Wistar rats, Toxicology International, 18, 2011, 130-6.
[6] D. Burukoglu, C. Baycu, F. Taplamacioglu, E.Sahin, and E. Bektur, Effects of nonsteroidal anti- inflammatory meloxicam on stomach, kidney, and liver of rats. Toxicology Indian Health, 2014, pii: 0748233714538484.
[7] AM. Khan, and S. Rampal, Effects of repeated oral administration of pazufloxacin mesylate and meloxicam on the antioxidant status in rabbits, Journal of American Association For Laboratory Animal Science. , 53(4) ,2014, 399-403.
[8] H. Li, M. Hortmann, A. Daiber, M. Oelze, MA. Ostad, PM. Schwarz, H. Xu, N. Xia, AL. Kleschyov, C. Mang, A. Warnholtz, T. Munzel,and U. Forstermann , Cyclooxygenase 2-selective and nonselective nonsteroidal antiinflammatory drugs induce oxidative stress by upregulating vascular NADPH oxidases, Journal of Pharmacology and Experimental Therapeutics, 326, 2008, 745–753.
[9] MH. Hassan , M. Ghobara , GM. Abd-Allah, Modulator effects of meloxicam against doxorubicin-induced nephrotoxicity in mice. Journal of Biochemistry and Molecular Toxicology, 28(8) , 2014, 337-46.
[10] M. Edfawy, MH. Hassan, A. Mansour, AA. Hamed,and HA. Amin, Meloxicam modulates oxidative stress status, inhibits prostaglandin E2, and abrogates apoptosis in carbon tetrachloride-induced rat hepatic injury, International Journal Toxicology, 31, 012, 276–286.
[11] SJ. Padayatty, A. Katz, Y. Wang, P. Eck, O. Kwon, JH. Lee, S. Chen, C. Corpe, A. Dutta, SK. Dutta , and M. Levine, Vitamin C as an antioxidant: evaluation of its role in disease prevention. The Journal Of the American College Of Nutrtion,22(1) , 2003,18-35.
[12] MS. Cooke, MD. Evans, ID. Podmore, KE. Podmore, KE. Herbert, N .Mistry, P. Mistry, PT. Hickenbotham, A. Hussieni, HR. Griffiths,and J. Lunec Novel repair action of Vitamin C upon in vivo oxidative DNA damage. FEBS Letters. ,363, 1998, 363–367.

[13] AG. Luna, Manaul of histological staining methods of the Armed Force Institute of Pathology. 3rd ed. (London: Mc Graw Hill Book Co,1968).
[14] V. N. Kartha, and S. Krishnamurthy, Factors affecting in vitro lipid peroxidation of rat brain homogenate. Indian journal of physiology and pharmacology ,22(1) , 1977, 44-52.
[15] A. Ayala, M. Muñoz, and S. Argüelles, Lipid Peroxidation: Production, Metabolism, and Signaling Mechanisms of Malondialdehyde and 4-Hydroxy-2-Nonenal. Oxidative Medicine and Cellular Longevity, 2014: 360438 , 2014, 31 pages .
[16] I. Villegas, C. Alarcón de la Lastra, C. La Casa, V, Motilva, and MJ. Martín, Effect of food intake and oxidative stress on intestinal lesions caused by meloxicam and piroxicam in rats. European Journal Pharmacology, 414, 2001, 79–86.
[17] C. Leeuwenburgh, and JW. Heinecke, Oxidative stress and antioxidants in exercise. Current Medicinal Chemistry,8(7) , 2001, 829–38.
[18] G.R. Buettner, The pecking order of free radicals and antioxidants: lipid peroxidation, α-tocopherol, and ascorbate. Archives of Biochemistry and Biophysics, 300, 1993, 535–43 .

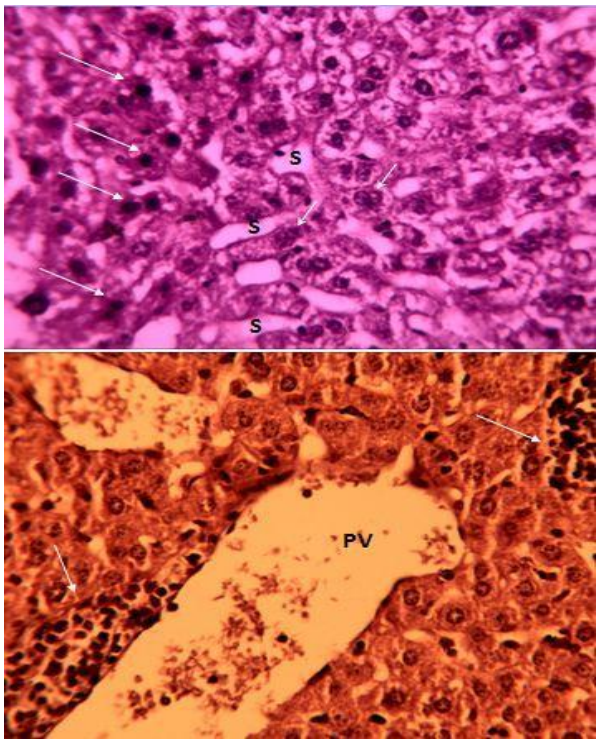
**Table(1): Biochemical results in mice liver**

| Groups | Doses Mg/kg bw | MDA Nmol/g tissue | ALT U/L | AST U/L | ALP U/L |
|---|---|---|---|---|---|
| control | - | $5.25\pm 1.53^{a}$ | $32.89\pm0.77^{a}$ | $85.66\pm4.10^{a}$ | $35.66\pm1.22^{a}$ |
| Vitamin c | 350 | $4.49\pm1.02^{a}$ | $44.34\pm0.84^{a}$ | $80.34\pm3.89^{a}$ | $42.32\pm2.13^{a}$ |
| Meloxicam | 0.4 | $15.51\pm2.82^{c}$ | $140.22\pm2.74^{b}$ | $230.22\pm3.11^{c}$ | $65.34\pm3.11^{b}$ |
| Vit+Meloxicam | 350+0.4 | $7.33\pm1.92^{b}$ | $54.19\pm0.86^{a}$ | $102.32\pm5.15^{b}$ | $39.01\pm2.02^{a}$ |

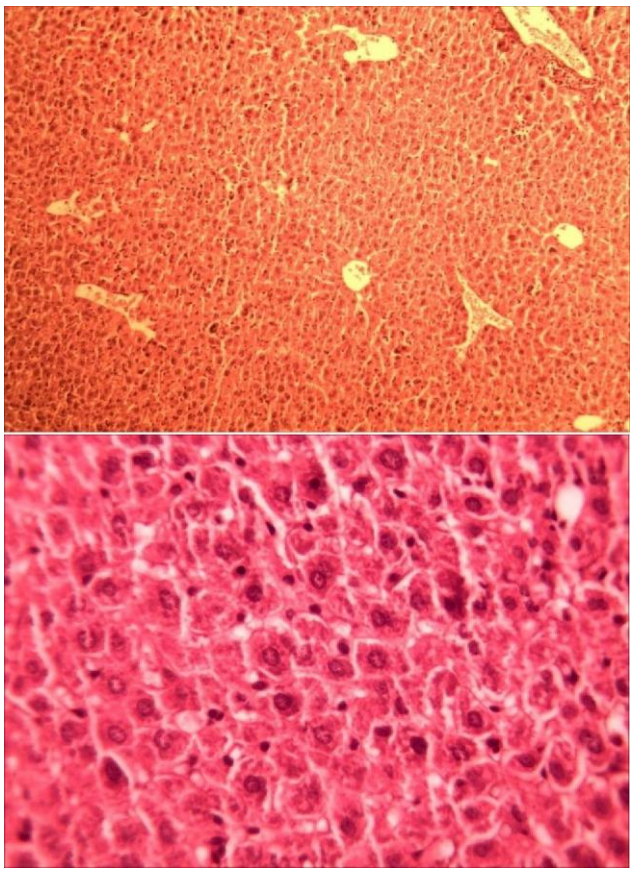
**Different letters were considered as significant, while similar letters were considered as non significant at ($p\leq 0.01$)**



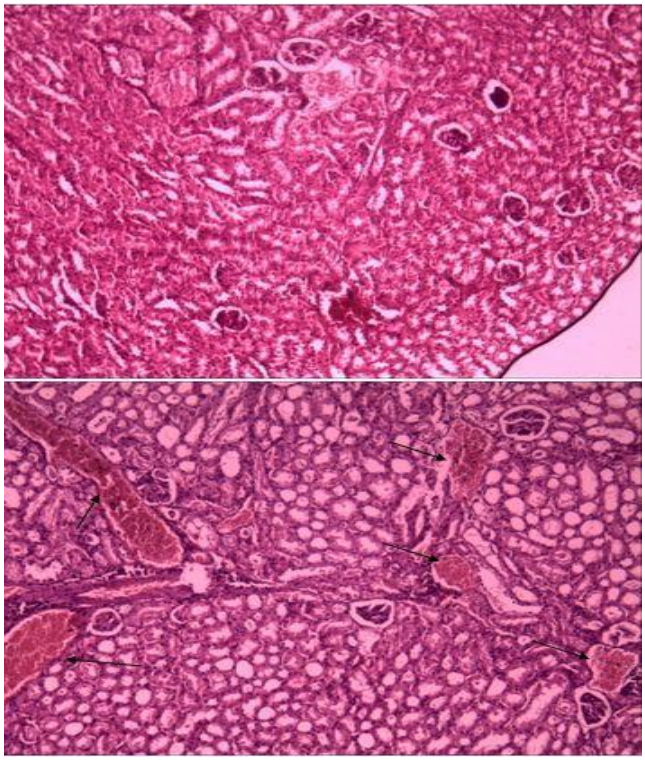

Fig(1): Sections through the liver of control group, normal histological architecture,central vein(CV), healthy hepatocytes(arrow) and normal sinusoidal lumen, up:100X, down: 400x,H&E



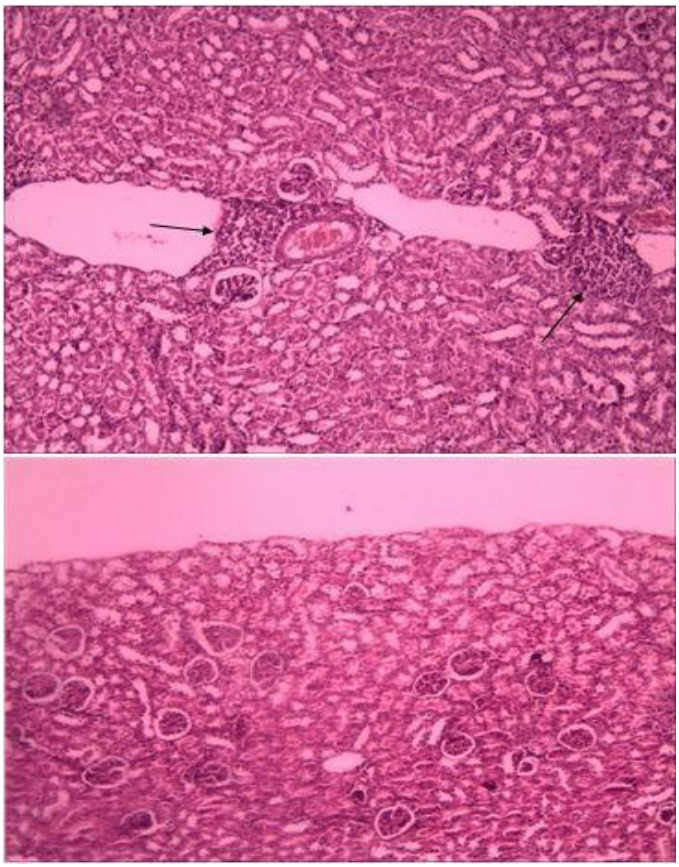

Fig(2): Sections through meloxicam treated mice liver showing: up) dilation in blood sinusoid lumen (S), degeneration of hepatocytes, mainly necrosis (arrows), down) inflammatory cells aggregation (arrow) near the portal vein(PV),400X, H&E



Fig(3): Sections through meloxicam plus vitamin C treated mice liver showing approximately normal histological structure: up) 100X , down) 400X, H&E



Fig(4): Sections through mice kidney showing: up) Normal histological structure in control group,100X, : down) congestion of blood vessels (arrows) with blood cells in meloxicam treated group 400X, H&E



Fig(5): Sections through mice kidney showing: up) meloxicam treated group having degeneration of kidney tubules lining cells and inflammatory leucocytes infiltration (arrows), down) Approximately normal structure of kidney in meloxicam plus vitamin group, 100X, H&E