

# Hepatotoxic Evaluation of Carboxymethylcellulose in Adult Wistar Rats

Isa AS<sup>1</sup>, Muhammad MS<sup>1</sup>, Hudu AA<sup>1</sup>, Jamba BI<sup>1</sup>, Magaji MG<sup>2</sup>

<sup>1</sup>Department of Human Physiology, Faculty of Basic Medical Sciences, College of Medical Sciences, Ahmadu Bello University, Zaria, Kaduna Nigeria.

<sup>2</sup>Department of Pharmacology and Therapeutics, Faculty of Pharmaceutical Sciences, Ahmadu Bello University Zaria Kaduna, Nigeria.

\*Correspondence: Email: isa.s.sherif@gmail.com

Submitted: 2 September, 2021. Published: July 2022

## ABSTRACT

**Background:** Carboxymethylcellulose (CMC) is an essential derivative of cellulose, which is employed in various applications in the food industry, detergents, cosmetics, pharmaceuticals, textiles, paper, adhesives as well as ceramics. There is a current concern that Carboxymethylcellulose may have some biological activity. This study evaluated the effects of CMC on some liver indices of adult Wistar rats.

**Methodology:** Twenty (20) adult Wistar rats divided into four groups I-IV of five animals each were used in the study. Group one (control) received distilled water, while groups II-IV received 5 mg/ml, 10 mg/ml, and 20 mg/ml respectively. Blood was collected and processed for laboratory assessments of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), albumin and total protein. **Results:** The results obtained showed that; CMC had no significant effects on albumin, total protein, and alanine aminotransferase (ALT). In contrast, a significant increase was seen in aspartate aminotransferase (AST) level, which was dose dependent. The result also showed a significant decrease in alkaline-phosphatase (ALP) level. **Conclusion:** Our findings suggest that CMC possesses no hepatotoxic properties and the changes seen in our results are within the physiological range

**Keywords:** Carboxymethylcellulose, Liver enzymes, Hepatotoxicity

## INTRODUCTION

Cellulose is a high molecular weight environmentally friendly and biodegradable material<sup>[17]</sup> commonly converted into useful derivatives<sup>[6,21]</sup>. The applicability of cellulose requires an alternative



Access to the article

Website: [www.jmbsr.com.ng](http://www.jmbsr.com.ng)

doi: [10.5281/zenodo.6457945](https://doi.org/10.5281/zenodo.6457945)

### How to cite this article

Isa AS, Muhammad MS, Hudu AA, Jamba BI, Magaji MG .  
Hepatotoxic Evaluation of Carboxymethylcellulose in Adult  
Wistar Rats . J Med & Bas Sci  
Res. 2022;3(2):33-40. DOI: 10.5281/zenodo.6457945

For Reprint: [info.jmbsr@gmail.com](mailto:info.jmbsr@gmail.com)

J Med & Bas Sci Res. Vol. 3, No. 2, 2022.

pathway that converts cellulose to its derivatives such as carboxymethylcellulose <sup>[10,6,27]</sup>. Carboxymethylcellulose (CMC) is derived from cellulose alongside other essential derivatives. Sodium carboxymethyl cellulose is the most typical form of CMC obtained from the reaction of cellulose with sodium mono-chloroacetate (MCA) in an alkaline medium through etherification process <sup>[3,16]</sup>. Among the cellulose ethers, carboxymethyl cellulose is the most water-soluble derivative which has immense importance for specific industrial applications used in a everyday life <sup>[5,7]</sup>.

CMC has been employed in everyday life in a wide variety of industrial and scientific processes due to its desirable qualities, which includes solubility, low toxicity <sup>[17]</sup>, cheap, bio-adhesion, cross-link to form hydrogels essential for tissue regeneration <sup>[9,18]</sup>, control release of drugs. Wang *et al.* <sup>[29]</sup> reported that sodium carboxymethyl cellulose improves the pharmacodynamics of 1-deoxyojirimycin by changing its absorption characteristics and pharmacokinetics in rats, among others. These desirable features give CMC its relevance in industrial applications in food products, cosmetics, pharmaceuticals, textiles, paper, in detergents and soaps, adhesives, and prints inks <sup>[4,14]</sup>.

CMC is also employed in intradermal drug delivery and drug formulations <sup>[6]</sup>. Many interests have been seen recently in the medical science regarding the use of CMC in specific ways that directly impact on human health including surgery, orthopaedic devices and drugs. The relevance of CMC in both food and pharmaceutical industries, has led to its increasing daily consumption especially because it is known to be biologically inert. There are limited studies on the biological activities of CMC, however, recently studies have

demonstrated that CMC possesses biological activities that includes pro-oxidative stress activity and effects on motor endurance and these has led to investigating the physiological properties of CMC <sup>[14,30]</sup>. This study investigated the hepatotoxic effects of orally administered CMC in adult male Wistar rats

## MATERIALS AND METHODS

Twenty (20) adult male albino Wistar rats, carboxymethylcellulose (CMC), plastic cages, feeders, sawdust, animal feed, water, weighing balance, needles and syringes, oral cannulas, dissecting kits, test tubes, ketamine, diazepam, and cotton wool.

### Chemicals/Reagents

Carboxymethyl cellulose (CMC) (product number: 27929, BDH Chemicals Ltd., Poole, England), was donated by Ahmadu Bello University, Zaria, Department of Pharmacology and Therapeutics, Ahmadu Bello University. All other chemicals used were analytically graded.

### Animals

Twenty (20) adult male Wistar rats with an average body weight of 150 g were used in the study. The animals were housed in plastic cages and allowed access to food and water *ad libitum*. The animals were allowed to acclimatise for two weeks before the commencement of the experiment. All experimental protocols were executed in accordance with the Ahmadu Bello University research policy and ethic and regulations governing the care and use of experimental animals (NIH publication number 85-23, 2010).

### Animal Groupings

The animals were divided into four (4) groups.

Group I: received distilled water based on their body weight orally.

Group II: received CMC 5 mg/ml per body weight orally.

Group III: received CMC 10 mg/ml per body weight orally

Group IV: received CMC 20 mg/ml per body weight orally.

All administrations were done between 09:00 and 10:00 hours each day for two (2) weeks.

### Animal sacrifice and blood collection

After two (2) weeks of the experiment, the rats were anaesthetised using ketamine (75 mg/kg) and diazepam (5 mg/kg), after which the blood samples were collected by cardiac puncture and transferred into plain test tubes using 5ml syringe. Serum samples were separated by centrifugation and stored until the biochemical assay was done.

### Assessments of Liver function Parameters

The serum total protein and albumin content were assayed using modified Biuret and Dumas method [8,25]. Alkaline phosphatase (ALP) activity was determined by the method of Ahmed and King [1], while the activities of aspartate aminotransferases and alanine aminotransferases (AST and ALT respectively) were determined by the method of Reitman and Frankel [24]. The assay kits used were obtained from Randox Laboratories, Co-Atrim, UK

### Statistical analysis

Results obtained were analysed using the Statistical Product and Service Solutions (SPSS version 17.0). All data were analysed using one-way analysis of variance (ANOVA), followed by

Tukey's *Post-hoc* test for multiple comparisons. Results were expressed as Mean  $\pm$  SEM,  $P < 0.05$  was taken as an acceptable level of significant difference between groups.

## RESULTS

### Alanine aminotransferase (ALT)

The result of ALT is represented in figure 1. The mean ALT level for the control group was  $7.8 \pm 0.86$  IU/L and that of the CMC groups was  $8.8 \pm 0.58$  IU/L,  $8.8 \pm 1.16$  IU/L and  $8 \pm 0.71$  IU/L for 5, 10 and 20 mg/ml respectively. There was no statistically significant difference ( $P > 0.05$ ) in alanine aminotransferase (ALT) level between the control and CMC groups.

### Aspartate aminotransferase (AST)

The result of AST is represented in figure 2. The mean AST level for the control group was  $13.6 \pm 1.57$  IU/L and that of the CMC groups (5, 10, 20 mg/ml) were  $26.4 \pm 2.21$  IU/L,  $26.8 \pm 2.71$  IU/L and  $25 \pm 1.92$  IU/L respectively. Aspartate aminotransferase (AST) was significantly higher ( $P < 0.05$ ) in CMC groups as compared to control.

### Alkaline phosphatase (ALP)

The result of ALP is represented in figure 3. The mean ALP level for the control group was  $79.23 \pm 9.88$  IU/L and that of the CMC groups were  $59.25 \pm 7.92$  IU/L,  $46.42 \pm 3.07$  IU/L and  $46.57 \pm 9.21$  IU/L respectively. Alkaline phosphatase (ALP) concentration was significantly decreased ( $P < 0.05$ ) in the CMC groups compared to the control.

### Albumin

The result of Albumin is represented in figure 4. The mean Albumin level for the control group

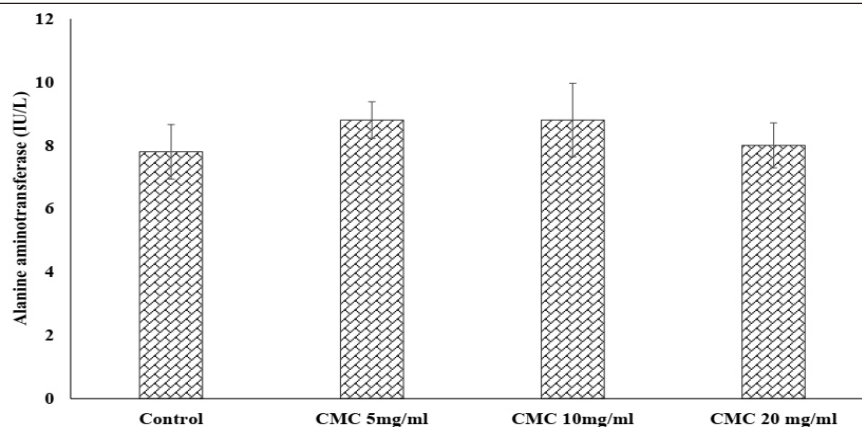


Figure 1: Effects of CMC on alanine aminotransferase level in male albino Wistar rats.

Data is shown as Mean ± SEM

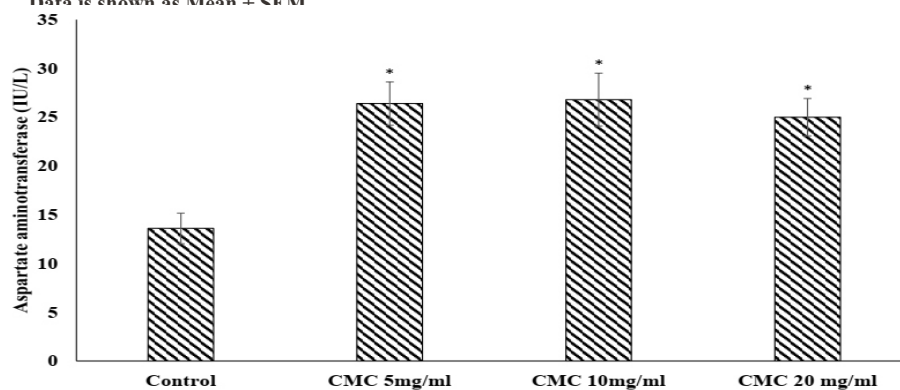


Figure 2: Effect of CMC on Aspartate aminotransferase level in male albino Wistar rats. Data is shown as Mean ± SEM, mean values with (\*) have significant differences ( $P < 0.05$ ) when compared to the control group.

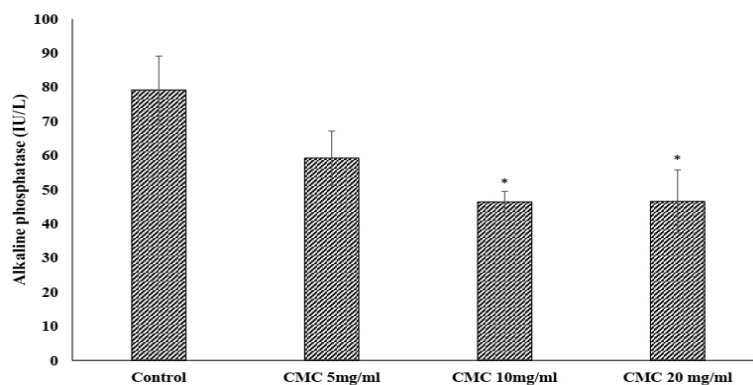


Figure 3: Effect of CMC on Alkaline phosphatase concentration in male albino Wistar rats. Data is shown as Mean ± SEM, mean values with (\*) have significant differences ( $P < 0.05$ ) when compared with the control.

was  $3.06 \pm 0.16$  g/dl and that of the CMC groups were  $3.13 \pm 0.07$  g/dl,  $2.83 \pm 0.19$  g/dl and  $2.77 \pm 0.13$  g/dl respectively. There was no statistically significant difference ( $P < 0.05$ ) in Albumin level between the control and CMC groups.

### Total Proteins

The result for Total protein is represented in figure 5. The mean total protein level for the control group was  $7.59 \pm 0.38$  g/dl, and that of the CMC groups were  $7.79 \pm 0.43$  g/dl,  $7.09 \pm 0.1$  g/dl and  $8.08 \pm 0.43$  g/dl respectively. There was no statistically significant difference ( $P < 0.05$ ) in Total proteins level between the control and CMC groups.

## DISCUSSION

Carboxymethyl cellulose (CMC) has many desirable applications, such as the formation of pharmaceutical products, cosmetics, food additives, agricultural chemicals etc. Therefore, it is extensively consumed daily, either directly or indirectly. This study examined the effects of treatment of rats with varying doses of CMC on serum liver enzyme concentration of alanine aminotransferase (ALT), aspartate aminotransferase (AST) alkaline phosphatase (ALP) and total protein, albumin, in a bid to evaluate the hepatotoxicity of CMC. The serum level of certain liver enzymes like Aspartate aminotransferase (AST), alanine aminotransferase (ALT),  $\gamma$ -glutamyl transferase (GGT) and alkaline phosphate (ALP), are useful indicators of liver injury<sup>[15]</sup>. Serum levels of ALT and AST are elevated in blood during tissue damage, especially liver cell damage. Thus, these enzymes possess a direct proportional relationship to liver tissue damage<sup>[31]</sup>. These hepatic enzymes may highlight the presence of specific or class of

diseases<sup>[15,21]</sup>. Naturally, alcoholic and non-alcoholic fatty liver hepatocellular damage is associated with an elevated serum level of GGT<sup>[13,23]</sup> and ALT<sup>[15]</sup> respectively. The magnitude of hepatocellular injury is indicated by elevated serum level of ALT and AST<sup>[22]</sup>. Elevated serum concentrations of ALP and GGT signify an impaired bile flow or cholestasis. The ratio of AST/ALT has been reported in literature as an index used in differentiating alcoholic from non-alcoholic fatty liver disease<sup>[13]</sup>. Elevated ALP may indicate problems not only related to the liver, but also gall bladder, bone, kidney tumours, and infections<sup>[13]</sup>. In our study, we observed no significant changes in the ALT recorded in the groups of animals that were treated with CMC compared to the control group. This suggests that the integrity and normal functioning of the liver may not have been compromised by the administration of CMC. The results obtained also showed a significant decrease in the level of alkaline phosphatase (ALP) in the groups of animals treated with carboxymethyl cellulose (CMC) compared to the control group. The reduction in ALP activity could be due to the inhibition of the enzyme's activities as it is in the case with the consumption of polluted *Asa* water<sup>[20]</sup> or metabisulphite<sup>[2]</sup>. The significant change seen in ALP might not arise from the liver, this is because, ALP is also found in other organs including the bone (in higher concentration), placenta, kidney and intestine<sup>[27]</sup>. Thus, ALP is a less specific biomarker for hepatotoxicity. The results obtained also showed a significant increase in the level of Aspartate aminotransferase (AST) in the groups of animals treated with carboxymethyl cellulose (CMC) compared to the control group. The increased AST level might be due to injuries to other body tissues or organs other than the liver

itself because AST is found in highest concentration in the heart compared to other tissues of the body such as liver, skeletal muscle and kidney [26]. Thus, AST is also considered a less specific biomarker enzyme for hepatocellular injury, as it can also signify abnormalities in heart, muscle, brain or kidney. Elevation in the activities of serum AST as observed in this study may have been due to leakage from the organs into extracellular fluids due to change in endothelial permeability [12]. Normally, the enzyme will not always be found in the serum, except there is damage to one or more organs or tissues of the body. Therefore, enzymes from diseased tissues or organs (e.g., cardiac, hepatic, and neoplastic diseases) and drug assault or other xenobiotics may have been manifested in the serum resulting in increased activity since they must have leaked from the diseased tissue [19]. However, despite the significant increase in AST in our study, the results may not be indicative of tissue damage because they are within the normal range for Wistar rats which is 5-153 IU/L [32,33]. The results obtained also showed no significant changes in the activities of Albumin and total protein, further corroborating the results that CMC may not be hepatotoxic. Taken together our study indicates that the CMC does not possess hepatotoxic properties

## CONCLUSION

We conclude based on our findings that CMC may not have hepatotoxic potential and the increase in AST observed in this study is within the acceptable normal range for Wistar rats.

## Conflict of interest

We declare no conflict of interest

## REFERENCES

1. Ahmed Z, King, E. J. Placenta phosphatases. *Biochemistry. Biophys. Acta.* 1959;34:313-15.
2. Akanji MA, Olagoke OA, Oloyede OB. Effect of chronic consumption of metabisulphite on the integrity of the rat kidney cellular system. *Toxicology.* 1993;81(3):173-9.
3. Asl SA, Mousavi M, Labbafi M. Synthesis and characterisation of carboxymethyl cellulose from sugarcane bagasse. *Journal of Food Processing Technology.* 2017; 8(8):1-5.
4. Aslam R, Mobin M, Aslam J, Lgaz H, Chung IM. Inhibitory effect of sodium carboxymethylcellulose and synergistic biodegradable gemini surfactants as effective inhibitors for MS corrosion in 1 M HCl. *Journal of Materials Research and Technology.* 2019;8(5):4521-33.
5. Bono A, Ying PH, Yan FY, Muei CL, Sarbatly R, Krishnaiah D. Synthesis and characterisation of carboxymethyl cellulose from palm kernel cake. *Advances in Natural and Applied Sciences.* 2009;3(1): 5-11.
6. Brandley A, Hollfelder R, Nesaei S, VanWie B, Abu-Lail N, Gozen BA. Direct-ink-writing of degradable carboxymethylcellulose. *Procedia Manufacturing.* 2018;26:993-1002.
7. dos Santos DM, de Lacerda Bukzem A, Ascheri DP, Signini R, de Aquino GL. Microwave-assisted carboxymethylation of cellulose extracted from brewer's spent grain. *Carbohydrate polymers.* 2015;131:125-33..
8. Dumas JB. *Procedes de l'analyse organique.* Ann. Chim. Phys. 1831;47:198-205.
9. Dutta SD, Patel DK, Lim KT. Functional

- cellulose-based hydrogels as extracellular matrices for tissue engineering. *Journal of biological engineering*. 2019;13(1):1-9.
10. Haleem N, Arshad M, Shahid M, Tahir MA. Synthesis of carboxymethyl cellulose from waste of cotton ginning industry. *Carbohydrate polymers*. 2014;113:249-55.
  11. Huang JF, Ou YX, Li XM, Lin ZC, Leng B, Xue Y, Su Y, Pan YT. The preventive effect of cellulose from *Pleurotus eryngii* on fatty liver of hyperlipidemic rats. *Biomedical Research*. 2018;29(9).
  12. Huang TH, Lii CK, Chou MY, Kao CT. Lactate dehydrogenase leakage of hepatocytes with AH26 and AH Plus sealer treatments. *Journal of endodontics*. 2000;26(9):509-11.
  13. Iluz-Freundlich D, Zhang M, Uhanova J, Minuk GY. The relative expression of hepatocellular and cholestatic liver enzymes in adult patients with liver disease. *Annals of hepatology*. 2020;19(2):204-8.
  14. Isa AS, Muhammad MS, Hudu AA, Jamba BI, Choji ES, Isah HO, Magaji MG. Assessment of cognitive and motor endurance activities in male wistar rats administered carboxymethyl cellulose. *African Journal of Biomedical Research*. 2019;22(2):195-9.
  15. Jain RB. Concentration of selected liver enzymes across the stages of glomerular function: the associations with PFOA and PFOS. *Heliyon*. 2019;5(7):e02168.
  16. Kinate B, Cornelius W. Preparation and characterization of carboxymethyl cellulose (CMC) from cassava peels, irish potatoes, yam peels. *Am. J. Eng. Res*. 2019;8(3):133-7.
  17. Lan W, He L, Liu Y. Preparation and properties of sodium carboxymethyl cellulose/sodium alginate/chitosan composite film. *Coatings*. 2018;8(8):291.
  18. Malik NS, Ahmad M, Minhas MU. Cross-linked  $\beta$ -cyclodextrin and carboxymethyl cellulose hydrogels for controlled drug delivery of acyclovir. *PloS one*. 2017;12(2):e0172727.
  19. Oloyede OB, Sunmonu TO. Decrease in activities of selected rat liver enzymes following consumption of chemical effluent. *Journal of Applied Sciences and Environmental Management*. 2008;12(3).
  20. Oloyede OB, Sunmonu TO, Adeyemi O, Bakare AA. A biochemical assessment of the effects of polluted water from Asa river in rat kidney. *Journal of Biochemistry and Molecular Biology*. 2003;17:25-32.
  21. Qin G, Lu L, Xiao Y, Zhu Y, Pan W, Xu X, Shen S, Das UN. A cross-sectional study of the relationship between serum liver enzymes level and the incidence of impaired fasting glucose in males and females. *Medical science monitor: international medical journal of experimental and clinical research*. 2014;20:1319.
  22. Rachtanapun P. Blended films of carboxymethyl cellulose from papaya peel (CMCp) and corn starch. *Agriculture and Natural Resources*. 2009;43(5):259-66.
  23. Rajabian M, Hussein Nejad E, Taghizade Moghaddam H. Comparison of Elevated Liver Enzymes in Type 2 Diabetic Patients in User and Non-User of Statin. *International Journal of Pediatrics*. 2018;6(12):8757-64.
  24. Reitman S, Frankel S. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *American journal of clinical pathology*. 1957;28(1):56-63.
  25. Savory J, Pu PH, Sunderman Jr FW. A biuret

- method for determination of protein in normal urine. *Clinical chemistry*. 1968;14(12):1160-71.
26. Gowda S, Desai PB, Hull VV, Math AA, Vernekar SN, Kulkarni SS. A review on laboratory liver function tests. *The Pan African Medical Journal*. 2009;3.
27. Singh A, Bhat TK, Sharma OP. Clinical Biochemistry of Hepatotoxicity. *J Clinic Toxicol*. 2011;S4:001. doi: 10.4172/2161-0495.
28. Singh RK, Singh AK. Optimization of reaction conditions for preparing carboxymethyl cellulose from corn cobic agricultural waste. *Waste and Biomass Valorization*. 2013;4(1):129-37.
29. Wang L, Peng J, Wang X, Zhu X, Cheng B, Gao J, Jiang M, Bai G, Hou Y. Carboxymethylcellulose sodium improves the pharmacodynamics of 1-deoxynojirimycin by changing its absorption characteristics and pharmacokinetics in rats. *Die Pharmazie-An International Journal of Pharmaceutical Sciences*. 2012;67(2):168-73.
30. Muhammad MS, Magaji RA, Mohammed A, Isa AS, Magaji MG. Effect of resveratrol and environmental enrichment on biomarkers of oxidative stress in young healthy mice. *Metabolic brain disease*. 2017;32(1): 163-70.
31. Hasan KM, Tamanna N, Haque MA. Biochemical and histopathological profiling of Wistar rat treated with Brassica napus as a supplementary feed. *Food science and human wellness*. 2018;7(1):77-82.
32. Boehm O, Zur B, Koch A, Tran N, Freyenhagen R, Hartmann M, Zacharowski K. Clinical chemistry reference database for Wistar rats and C57/BL6 mice. (2007): 547-554.
33. Riris ID. methoxy bergenin's effect on total protein, albumin and globulin levels of wistar rats'blood. *Jurnal Der Pharma Chemica*. 2017;9(13):90-94.