



Histomorphological Effects of Tadalafil on the Liver of Adult Wistar Rat

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ABSTRACT: Tadalafil is used to treat erectile dysfunction and improve exercise capacity in people with pulmonary arterial hypertension. It promotes blood circulation to the penis during sexual stimulation, thus facilitating the occurrence of an erection. This study examined tadalafil effects on the liver, focusing on potential hepatotoxicity and liver function adverse effects. It aims to contribute to knowledge on tadalafil misuse and abuse, promoting informed decision-making and raising awareness. The study was conducted using fifteen (15) adult Wistar rats randomly assigned into three groups consisting of 5 rats each. Group A was the control group while group B (10 mg/kg of tadalafil) and C (20 mg/kg of tadalafil) were the low dose and high dose groups respectively. Oral administration of tadalafil in treated rats showed evidence of vascular congestion and dilation in the liver tissue, at 20 mg/kg there was periportal hepatitis. Assay of liver function parameters showed increase in liver enzymes across treatment groups compared to control. The presence of periportal hepatitis indicates potential inflammation in the region surrounding the portal area of the liver which suggests that tadalafil may have an impact on hepatocyte morphology and may also influence the inflammatory response in the periportal region of the liver. Hence, Tadalafil should be taken with caution.

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Tadalafil belongs to a selective class of phosphodiesterase-5 inhibitors used in the treatment of erectile dysfunction (ED), arterial hypertension (PAH), and benign prostatic hypertrophy. The Food and Drug Administration (FDA) initially authorized its usage in 2003 for ED (Briganti *et al.*, 2005). Tadalafil exhibits a higher preference for targeting PDE5 compared to sildenafil and other PDE5 inhibitors, which boost a longer duration of action. These characteristics have rendered it a more viable choice for convenient administration in the management of PAH (Gaines, 2004). The liver which is one of the largest organs in the body has numerous roles which includes bile production, Fat-Soluble Vitamin Storage and Metabolism, Drug Metabolism (O'Brien *et al.*, 2015). The liver, spleen, and bone marrow are a few of the organs throughout the body where hemolysis occurs (Almazroo *et al.*, 2017). The key location for drug metabolism is the liver, through

which the majority of medications must travel (Wespes *et al.*, 2013). Within the liver, enzymes serve to either convert active medications into inactive states or activate prodrugs into their active metabolites. This liver's primary system for drug metabolism is comprised of a specific group of cytochrome P-450 enzymes. The rate at which various medications undergo metabolism hinges on the abundance of cytochrome P-450 enzymes (Cotran *et al.*, 2005). An excess concentration of a drug in the bloodstream can lead to an overload of these enzymes, as their metabolic capacity is limited (Kumari *et al.*, 2016). The cytochrome P-450 enzymes are impacted by a variety of things, including food and medications. These compounds boost a drug's effects (including adverse effects) if they make it more difficult for the enzymes to break it down (Rajfer *et al.*, 2007). The effects of a medicine are diminished if the components make it easier for enzymes to break down that drug

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(Krause *et al.*, 1992). This study seeks to investigate the histomorphological effects of tadalafil on the liver of adult Wistar rat.

MATERIALS AND METHODS

The study was conceptualized in the department of human anatomy and cell biology. Approval was obtained from the Research and Ethics Committee of the Department of Human Anatomy and Cell Biology, Delta State University DELSU/CHS/ANA/2023/10.

The study comprised of 15 adult Wistar rats which were grouped into 3 groups with 5 rats each; high dose, low dose and control group respectively. The rats were kept inside an iron cage with compartments. The cages were well ventilated and kept under controlled environmental conditions of temperature ($25\pm 5^{\circ}\text{C}$), relative humidity ($50\pm 5\%$) and 12-hour light / dark cycle. They were fed everyday with standard grower's mash (feed) and had free access to water. The drug was administered for 6 weeks (42 days), following which the rats underwent an overnight fast and were euthanized by cervical dislocation. Subsequently, their livers were harvested to identify any histopathological and morphological changes. Blood samples were collected by placing the rat in a supine position, sterilizing the abdominal area, making a midline incision, locating blood vessels, and collecting the required volume of blood. The liver was then fixed in 10% formal saline and subjected to liver function testing. Parameters used for the liver function test includes;

Total protein which is one of the biochemical parameters used to measure the liver function is a test that measures the overall protein content in the blood, including albumin and globulins. Abnormal levels may indicate liver disease or malnutrition. The liver and gills total protein is estimated using the method of Tietz (Pric Bossuyt, and Bruns, 1976).

Alanine transaminase (ALT) is an enzyme primarily located within the liver, with smaller quantities also present in tissues such as the heart and kidneys. The assay for ALT activity in serum and tissue homogenates is carried out using the method of Reitman and Frankel (1957). The enzyme's reaction is alanine + α -ketoglutaric acid \rightarrow pyruvic acid + glutamic acid, which is then reacted with 2,4 dinitrophenylhydrazine to form the corresponding color hydrazine.

Alkaline phosphatase (ALP) activity is estimated spectrophotometrically by kinetic method using a commercial diagnostic kit. The enzyme hydrolyzes a colourless substrate of phenolphthalein

monophosphate, giving rise to phosphoric acid and phenolphthalein, which at alkaline pH values change into a pink color.

Aspartate transaminase (AST) is an enzyme found in the liver and other organs. Elevated AST levels can be a sign of liver damage but are less specific to the liver than ALT. The assay for AST activity is similar to that of ALT but with a 60-minute incubation time. The activity of the enzyme is extrapolated from the AST standard curve and expressed as unit/ml.

Statistical analysis: Data were analyzed using descriptive statistics, and the results were expressed as the mean and standard error of the mean. Differences in mean value were assessed and $p < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Rats in the control group showed that the liver tissue composed predominantly of the hepatic parenchyma and portal regions. The hepatocytes (H) appear polygonal and are disposed in sheet with a well outlined nucleus (N) the hepatocytes are separated by the sinusoids (S) with thin endothelial lining. The Portal Region (Circle), composed of branches of the Hepatic portal vessels (HPV), and bile duct (BD) appear normal. Section histomorphology appear apparently normal (Figure 1a and 1b).

Rats treated with 10 mg/kg of Tadalafil showed that the liver tissue composed predominantly of the hepatic parenchyma and portal regions. The hepatocytes (H) appear polygonal and are disposed in sheet with a well outlined nucleus (N). The hepatocytes are separated by the sinusoids (S) with thin endothelial lining and contained activated Kupffer cells (arrow head). The Portal Region (Circle) composed of branches of the Hepatic portal vessels (HPV), bile duct (BD) and showed vascular congestion. Section showed periportal hepatitis. (figure 2a & 2b)

Rats treated with 20 mg/kg of Tadalafil showed that the liver tissue composed predominantly of the hepatic parenchymal and portal regions. The hepatocytes (H) appear polygonal and are disposed in sheet with a well outlined nucleus (N). The hepatocytes are separated by the sinusoids (S) with thin endothelial lining and contained activated Kupffer cells (arrow head). The Portal Region (Circle), composed of branches of the Hepatic portal vessels (HPV), bile duct (BD) showed vascular congestion and dilated sinusoids. Section showed periportal hepatitis (Figure 3a and 3b).

The results obtained from the liver function test showed significant increase ($p=0.001$) in ALT, AST

and ALP of treated rats compared to control. Multiple comparison between groups using Tukey's test showed statistically significant increase ($p=0.001$) in the mean serum ALT level ($41.19\text{U/L}\pm 5.044$) of rats treated with 20 mg/kg of tadalafil compared to control ($22.98\text{U/L}\pm 5.044$). Also, there was significant increase in mean serum AST level ($40.93\text{U/L}\pm 1.919$) and ALP level ($225.50\text{U/L}\pm 17.410$) of rats treated with 20 mg/kg body weight of tadalafil compared to the control of each enzyme ($28.48\text{U/L}\pm 1.919$; $138.50\text{U/L}\pm 17.14$). More so, the changes observed were dose dependent as the mean serum concentration of the enzymes in treated rats that received 20 mg/kg with significantly ($P<0.05$) higher compared to the rats that received 10 mg/kg per body weight of tadalafil (Figure 4a). There was no significant ($p>0.05$) change in mean protein concentration in rats that received 10 mg/kg ($15.65\text{g/dl}\pm 1.815$) and 20 mg/kg ($12.48\text{g/dl}\pm 1.815$) per body weight of tadalafil (figure 4b) compared to the control ($15.94\text{g/dl}\pm 1.815$)

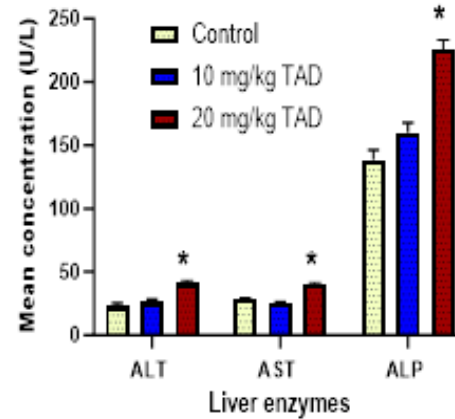


Fig 4a: Mean concentration of liver enzymes. Values are presented in graph as mean \pm SEM for each group; $n=5/\text{group}$, * $p<0.05$ denotes significant difference compared to control.

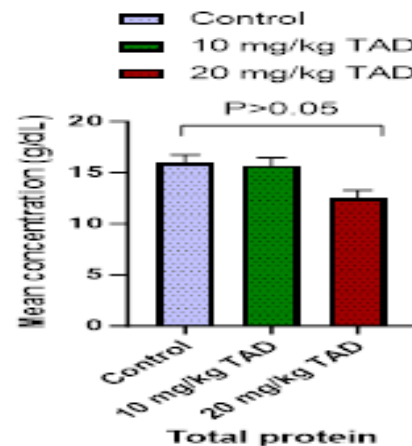


Fig 4b: Mean concentration of Total protein. Values are presented in graph as mean \pm SEM for each group; $n=5/\text{group}$, * $p<0.05$ denotes significant difference compared to control.

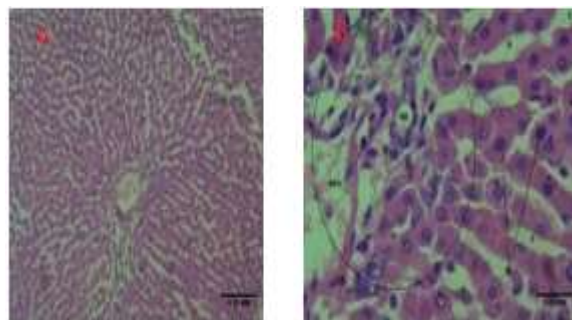


Fig 1a and 1b. The liver of adult Wistar rat in control group respectively ($\times 100$ & $\times 400$ respectively).

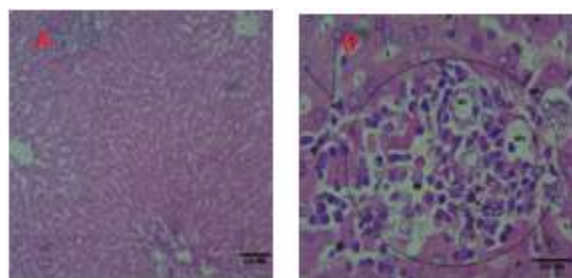


Fig 2a and 2b. The liver of adult wistar rat given 10 mg/kg of tadalafil respectively ($\times 100$ & $\times 400$ respectively).

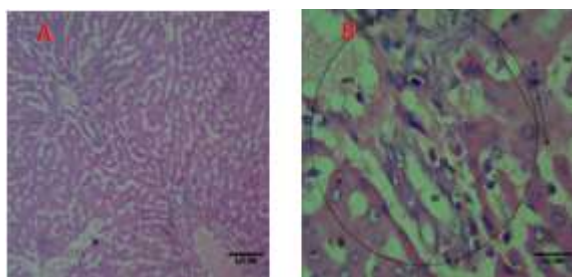


Fig 3a and 3b. The liver of adult Wistar rat given 20 mg/kg of tadalafil respectively ($\times 100$ & $\times 400$ respectively).

Tadalafil is metabolized predominantly in the liver by the cytochrome P450 enzyme system, specifically the CYP3A4 enzyme. This metabolism pathway is crucial in breaking down the medication into inactive compounds that can be eliminated from the body (Williams, 1972; Jelkmann, 2001).

The liver contains enzymes, notably cytochrome P450 enzymes, responsible for metabolizing various compounds (Sandroni, 2001). This present study found that rats across treatment groups showed hepatocytes which appeared polygonal and were arranged in sheets with well-defined nuclei. The hepatocytes were separated by the sinusoids with thin endothelial lining and contained activated Kupffer cells. The Portal Region, composed of branches of the Hepatic portal vessels and bile duct which showed vascular congestion and dilated sinusoids. The results

of this are consistent with impaired blood flow within the liver often seen in conditions such as congestive heart failure, cirrhosis, or portal hypertension. Tadalafil is metabolized predominantly in the liver by the cytochrome P450 enzyme system, specifically the CYP3A4 enzyme. This metabolism pathway is crucial in breaking down the medication into inactive compounds that can be eliminated from the body (Jelkmann, 2001).

The liver contains enzymes, notably cytochrome P450 enzymes, responsible for metabolizing various compounds (Sandroni, 2001). When aphrodisiacs or other substances enter the body, these enzymes work to break them down into forms that can be eliminated. Some aphrodisiacs could affect the activity of liver enzymes (Jakoby and Ziegler, 1990). They may inhibit or induce specific enzymes, altering the metabolism of other substances processed by these enzymes (Maton *et al.*, 1993). This interaction can impact the levels of medications or compounds in the body, affecting their effectiveness or causing unintended side effects (Lehmiller, 2017).

Herbal aphrodisiacs, derived from plants or herbs, often contain bioactive compounds that might interact with liver enzymes (Maton *et al.*, 1993). Individual responses to aphrodisiacs can vary due to factors like genetics, existing health conditions, medications taken, and overall liver health. Considering these variables is crucial when assessing how liver enzymes might respond to aphrodisiacs or any other substances: Alkaline phosphatase (ALP) and Aspartate aminotransferase (AST) are enzymes found in the liver; elevated levels can indicate liver damage or bile flow issues. Alanine aminotransferase (ALT) is another liver enzyme; increased levels suggest liver cell damage due to various conditions like hepatitis or cirrhosis. Total protein levels reflect liver function, with low levels signaling impaired function and high levels potentially indicating dehydration (Lehmiller, 2017).

However, administration of tadalafil has been confirmed to cause increase in ALP, AST and ALT levels in the level which can significantly affect tadalafil's clearance from the body. It was also discovered that tadalafil has no effect on the total protein level in the liver. According to a study conducted by Bektas *et al* (2016) to determine the effects of tadalafil and pentoxifylline on apoptosis and nitric oxide synthase in liver ischemia/reperfusion injury showed that there was an increase in the levels of ALT, AST, and ALP enzymes in both the 10 mg/kg and 2.5 mg/kg of tadalafil groups when compared to

the control group. This study suggests that tadalafil could negatively affect liver function at high doses.

Conclusion: Tadalafil administration might affect hepatocyte structure and inflammatory response in the periportal region. Moreso, vascular changes observed possibly linked to impaired blood flow may be associated with conditions like congestive heart failure, cirrhosis, or portal hypertension.

REFERENCES

- Almazroo, OA; Miah, MK; and Venkataramanan, R. (2017). Drug Metabolism in the Liver. *Clin Liver Dis.* 21 (1), 1-20. DOI: <https://doi.org/10.1016/j.cld.2016.08.001>
- Bektas, S; Karakaya, K; Can, M; Bahadir, B; Guven, B; Erdogan, N. and Ozdamar, S.O. (2016). "The effects of tadalafil and pentoxifylline on apoptosis and nitric oxide synthase in liver ischemia/reperfusion injury. *The Kaohsiung J. Med. Sci.* 32 (7), 339-347 DOI: <https://doi.org/10.1016/j.kjms.2016.05.005>
- Briganti, A; Salonia, A; Gallina, A; Saccà, A; Montorsi, P; Rigatti, P. and Montorsi, F. (2005). Drug insight: oral phosphodiesterase type 5 inhibitors for erectile dysfunction. *Nat. Clin. Pract. Urol.* 2 (5), 239-247. <https://doi.org/10.1038/ncpuro0186>
- Cotran, RS; Kumar, V; Stanley, L; Robbins, SL; Abbas, A. and Abul, K. (2005). Pathologic basis of disease. *Saunders.* 189 (1), 94-103. ISBN 07-216-75956
- Gaines, KK. (2004). Tadalafil (Cialis) and vardenafil (Levitra) recently approved drugs for erectile dysfunction. *Urol. Nur.* 24 (1), 46-48. PMID: 15054911
- Jakoby, WB; Ziegler, DM. (1990). The enzymes of detoxication. *J. Biol. Chem.* 265 (34):20715-8 doi:10.1016/S0021-9258(17)45272-0
- Jelkmann, W; (2001). The role of the liver in the production of thrombopoietin compared with erythropoietin. *Eur. J. Gastr. Hepat.* 13 :(7):791-801. doi:10.1097/00042737-200107000-00006
- Kumari, BS; Hanuja, GS; Nagabhusanam, MV; Reddy, DN; Bonthagarala, B. (2016). "Current Regulatory Requirements for Registration of Medicines, Compilation and Submission of Dossier in Australian Therapeutic goods

- Administration. *Int. J. Adv. Sci. Tech Res.* 2249-9954. ISSN 2249-9954
- Krause, D.S., Wolf, B.A. and Shaw, L.M., 1992. Acute aspirin overdose: mechanisms of toxicity. *Therap. drug monit.* 14: 441-441. DOI: 10.1097/00007691-199212000-00001
- Lehmiller, JJ. (2017). *The psychology of human sexuality* (Second ed.). Hoboken, NJ. ISBN 9781119164708.
- Maton, A; Jean Hopkins; Charles William McLaughlin; Susan Johnson; Maryanna Quon Warner; David LaHart; Jill D. Wright (1993). *Hum. Biol. Health*. Englewood Cliffs, New Jersey, USA: Prentice Hall. ISBN 978-0-13-981176-0. OCLC 32308337
- O'Brien, L; Hosick, PA; John, K; Stec, DE; Hinds TD. (2015) Biliverdin reductase isozymes in metabolism. *Tr. Endocrinol Metab.* 26 (4), 212-20. doi: 10.1016/j.tem.2015.02.001
- Pric Bossuyt, PM; Bruns, DE. (1976). "Tietz Fundamentals of Clinical Chemistry. *Med.* 3 (1).
- Rajfer, J; Aliotta, PJ; Steidle, CP; Fitch, WP; Zhao, Y; Yu, A. (2007). Tadalafil dosed once a day in men with erectile dysfunction: a randomized, double-blind, placebo-controlled study in the US. *Int. J. Imp Res.* 19, 1. DOI: <https://doi.org/10.1038/sj.ijir.3901496>
- Reitman, S; Frankel, S. (1957). "A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am J. Clin. Path.* 28 (1): 56-63. <https://doi.org/10.1093/ajcp/28.1.56>
- Sandroni, P; (2001). Aphrodisiacs past and present: A historical review. *Clin Auto Res.* 11: (5):303-307. doi:10.1007/bf02332975. ISSN 0959-9851
- Wespes, E; Amar, E; Eardley, I; Giuliano, F; Hatzichristou, D; Montorsi, F; Vardi, Y; Hatzimouratidis, K. (2013). Guidelines on male sexual dysfunction: erectile dysfunction and premature ejaculation. *Eur. Urol.* 57 (5), 804-814. doi: 10.1016/j.eururo.2010.02.020.
- Williams RT. (1972) Progress Report. Hepatic Metabolism of drugs. *Gut.* 13: 579-585. DOI: 10.1136/gut.13.7.579