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Histological Effect of Aloe Vera Gel on Liver of Albino Rats

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A B S T R A C T

The liver plays a vital role in metabolism, detoxification, and overall physiological homeostasis, making it susceptible to damage from both synthetic and natural compounds. Aloe vera, a commonly used medicinal plant, has gained attention for its anti-inflammatory, antioxidant, and hepatoprotective properties. This study aimed to assess the histological effects of orally administered Aloe vera gel on the liver of albino rats. Fifty (50) adult male and female albino rats were randomly divided into five groups (A–E) of ten rats each. Group A served as the control and received distilled water, while Groups B to E were administered Aloe vera gel orally at doses of 1.4 ml, 2.8 ml, 8.4 ml, and 13.9 ml for 14 days. Rats were weighed before and after treatment, and liver tissues were harvested for histological examination using standard hematoxylin and eosin staining protocols. Body weight data were analyzed using ANOVA with significance set at $p < 0.05$. All groups exhibited statistically significant ($p = 0.01$) increases in body weight post-treatment. Histologically, the control and low-dose groups (1.4 ml and 2.8 ml) showed normal liver architecture with preserved hepatic sinusoids and Kupffer cells. However, higher doses (8.4 ml and 13.9 ml) resulted in dose-dependent histopathological changes, including swollen hepatocytes, vascular congestion, central vein shrinkage, and hepatic necrosis. These findings indicate a potential hepatotoxic effect at elevated doses of Aloe vera gel. Caution is therefore advised in the therapeutic use of Aloe vera, especially at high concentrations or with prolonged exposure

INTRODUCTION

The liver is a vital organ in vertebrates responsible for numerous essential physiological functions, including such as bile production, nutrients' metabolism, waste products' removal, storage of glycogen and synthesis of plasma proteins (Farid et al., 2022). Its central role in drug metabolism and xenobiotic detoxification makes it particularly vulnerable to damage by toxic substances. With the increasing interest in herbal and alternative medicines, there has been a parallel need to understand the safety profiles and histological impacts of these natural products on critical organs such as the liver. Among the most widely used medicinal plants is Aloe vera, a succulent species belonging to the family Liliaceae. It has been extensively employed in traditional medicine and as a component of modern pharmaceuticals and cosmetics, due to its purported anti-inflammatory, antioxidant, and hepatoprotective properties.

Aloe vera is a desert plant that thrives in hot dry areas; and because of its ability to survive in certain severe climates, it has healing and antibacterial properties. Aloe vera gel is a juice product created from the inner pulp of Aloe vera leaves. Leaves of Aloe vera have been used to treat eye disorders as well as spleen and liver enlargements (Farid et al., 2022). Aloe vera has been found to have anti-inflammatory, wound healing, antidiabetic, anticancer, antibacterial, antiviral, purgative, oxytocic and lipid lowering properties (Madhav & Bairy, 2011). Furthermore, its antioxidant and anti-inflammatory actions have been shown to protect the liver. Aloe vera has seventy five therapeutically active elements like minerals, vitamin, enzyme, carbohydrate, lignin, saponin and salicylic acid (Farid et al., 2022).

LITERATURE REVIEW

Aloe vera has been extensively studied for both protective and potentially toxic effects on liver tissue in rats. Early investigations report hepatoprotective benefits: in an ischemia–reperfusion model, Wistar albino rats given 30 mg/kg Aloe vera gel for a month showed preserved liver architecture (minimal sinusoidal dilatation, reduced hemorrhage and vacuolization) compared to untreated controls, thanks to lowered oxidative stress and reduced iNOS expression (Sehitoglu et al., 2019). Similarly, in rats challenged with liver toxins such as malathion or cartap, Aloe vera notably improved antioxidant enzyme activity and attenuated histopathological injury (Gupta et al., 2023)

Conversely, some studies note hepatotoxicity. One pharmacovigilance study administering commercial Aloe vera "plus" gel to rats for up to 42 days reported elevated liver enzymes and histological signs of portal triaditis, vacuolization, hemorrhage, and necrosis (effects independent of dose but increasing over time) (Koroye et al., 2010). Moreover, chronic use of whole leaf extracts in Wistar rats produced moderate hepatic necrosis and portal inflammation even at sub acute doses (Nalimu et al., 2022). From the above, it is observed that moderate doses of purified inner gel appear hepatoprotective in specific injury models, whereas prolonged use of whole leaf or commercial products may induce liver damage. These findings reveal the need for thorough histological assessment research exploring Aloe vera's hepatic effects. This study therefore aims to investigate the histological effects of orally administered Aloe

vera gel on the liver tissue of albino rats. By employing standard histopathological techniques, the research will evaluate potential alterations in liver morphology and architecture

METHODOLOGY

Research Design

Fifty (50) male and female albino rats were used for this study. They were divided into five (5) equal groups (labeled A to E) of ten (10) rats each. Group A served as the control and the rats were given distilled water. Group B was administered 1.4ml Aloe Vera gel extract; Group C was administered 2.8ml Aloe Vera gel extract. Group D was administered 8.4ml Aloe Vera gel extract. Group E was administered 13.9ml Aloe Vera gel extract. The substance administration was given daily for 14 days (2 weeks) and the weights of both the test animal and control monitored every week. After the administration, the rats were put under light chloroform anesthesia and the liver harvested for histological processing. ANOVA was used to analyze the results of the weight and differences were considered significant at $P < 0.05$ level of confidence. All data are expressed as Mean ± Standard deviation. The results was presented in tables and comparisms made statistically.

Geographical Description of the Study Area

This study was carried out in the experimental site at the histology laboratory college of medicine, Ambrose Alli University Ekpoma, Edo State Ekpoma and the Irrua Specialist Teaching Hospital, Irrua, both in Edo state. Edo state lies between longitude 06o 04IE and 06o 43IE and latitude 05o 44IN and 07o 34IN with a land mass of 17, 450 sq.km located in the south-south geopolitical zone of Nigeria with a population of 3.1 million people (World Gazzetter, 2007).

Experimental Animals/Housing Condition

Fifty (50) Adult albino rats of comparable sizes and weights ranging from 90g to 130g (age of rats is five (5) weeks) was procured from the animal farm, college of medicine Ambrose Alli University Ekpoma and transferred to the experimental Laboratory at the histology laboratory college of medicine, Ambrose Alli University Ekpoma, Edo State, where they were allowed two (2) weeks of acclimatization. They were kept in wire mesh cages with tripod that separated the animal from its faeces to prevent contamination. During this period of acclimatization, the rats were fed with Growers' mash and water. The animals was maintained and utilized in accordance with the standard guide for the care and use of Laboratory animals.

Animal Grouping

The experimental animals were separated into five groups (A – E). Each group contains ten rats each (n = 10) using five (5) big cages to house them. Group A served as the control, while groups B - E served as the test groups.

Group B – E received doses of Aloe Vera gel. The dosages were prepared accordingly and weighed to determine the quantity to be administered.

Group A received only the normal feed (grower's mash) and water with no administration of Aloe Vera gel.

Study Duration

The preliminary studies, animal acclimatization, drug or substance procurement (dosage preparation and reconstitution), actual animal experiment and evaluation of results lasted for a period of 4 weeks (28 days). However, the actual administration of Aloe Vera gel to the test animals lasted for 2 weeks.

Substance of Study

Considerable amount of Aloe Vera was procured from the Ambrose Alli University Farm and stored at a temperature below 30oC in a cool place pending usage.

Substance Administration

The rats were weighed before the administration of the substances (Aloe Vera gel) and before they were sacrificed and similar weight measurements were done at the beginning and end of the experiment and the average weight recorded accordingly. The administration of the substance was via oral route thus:

1. Group A (Control) received only normal feed (growers' mash) and distilled water daily for 28 days (4 weeks).
2. Group B received 1.4ml of Aloe Vera gel, feed and distilled water daily for 14 days (2 weeks).
3. Group C received 2.8ml of Aloe Vera gel, feed and distilled water daily for 14 (2 weeks).
4. Group D received 8.4ml Aloe Vera gel, feed and distilled water daily for 14 days (2 weeks).
5. Group E received 13.9ml of Aloe Vera gel, feed and distilled water daily for 14 days (2 weeks).

Sample Collection and Analysis

Weight was measured before and after acclimatization and similar weight measurements was done at the end of each week and the average weight recorded accordingly. The liver of each rat was obtained at the end of the experiment under chloroform anaesthesia and fixed in 10% formalin for histological processing. The growth performance and feed utilization of the rats was determined at the end of the experiment as described by Dada and Ikuerowo (2009).

Histological Processing

The tissues was processed using automatic tissue processor according to the processing schedule used in Irrua Specialist Teaching Hospital, Edo State, Nigeria. The fixed plastic cassette tissues in 10% formalin was automatically processed by passing them through different grades of alcohol as follows:

70% alcohol	2hrs
80% alcohol	2hrs
90% alcohol	2hrs
90% alcohol	2hrs
95% alcohol	2hrs
Absolute	2hrs
Xylene 1	2hrs
Xylene II	2hrs
Molten paraffin wax 1	2hrs
Molten paraffin Wax II	2hrs

After the last timing, the tissues were removed from their plastic cassettes and placed at the centre of the metallic tissue mould and then filled with molten paraffin wax. They were also left to solidify after which they were placed in the refrigerator at 5oC for 15 minutes. After the blocks were left to cool in the refrigerator for the time stated above (15 minutes), the blocks were then removed from the metallic case using a knife and after which the paraffin wax at the side of the blocks were removed.

The blocks was then trimmed and cut serially at 5nm on a rotary microtome. The sections were floated in a water bath at 55oC and picked up by the use of clean frosted end slides. The frosted end slides were then placed on the hot plate for 40 minutes for adequate attachment of the sections on the slides after which the sections were de-waxed, hydrated, air dried and stored in a slide box ready for staining process.

Staining Procedure

Procedure

- De paraffinize the section: flame the slide on burner and place in the xylene. Repeat the treatment.
- Hydration: Hydrate the tissue section by passing through decreasing concentration of alcohol baths and water. (100%, 90%, 80%, 70%)
- Stain in hematoxylin for 3-5 minutes
- Wash in running tap water until sections "blue" for 5 minutes or less.
- Differentiate in 1% acid alcohol (1% HCl in 70% alcohol) for 5 minutes.
- Wash in running tap water until the sections are again blue by dipping in an alkaline solution (eg. ammonia water) followed by tap water wash.
- Stain in 1% Eosin Y for 10 minutes
- Wash in tap water for 1-5 minutes
- Dehydrate in increasing concentration of alcohols and clear in xylene
- Mount in mounting media
- Observe under microscope

(Ochei and Kolhatkar, 2000)

The slides were examined under a light microscope and photomicrographs were taken.

RESULTS

Table 1 shows the mean and standard deviation of initial and final body weight of adult wistar rats administered aloe vera gel.

The initial body weight of Group A (Control) was 93.13 ± 14.59, and the final body weight increased to 163.57 ± 33.72. The t-test value was -6.06, and the p-value of 0.01 indicates a significant change in body weight, suggesting that the Aloe Vera gel administered to the control group has a notable impact on their body weight increase.

For Group B (1.4 ml), the initial body weight was 82.53 ± 11.91, and the final body weight increased to 145.91 ± 25.58. With a t-value of -7.10 and a p-value of 0.01, the result is statistically significant. This implies that the administration of 1.4 ml of Aloe Vera gel caused a significant increase in body weight in the test subjects.

In Group C (2.8 ml), the initial body weight was 83.84 ± 20.92, which increased to 180.94 ± 30.02 by the end of the study. The t-test value was -8.39, and the p-value of 0.01 also shows a significant difference. This result indicates that the administration of 2.8 ml of Aloe Vera gel has a considerable effect on the body weight increase in this group.

For Group D (8.4 ml), the initial body weight of 80.02 ± 10.72 increased significantly to 130.74 ± 17.47. The t-value was -7.82, with a p-value of 0.01, which demonstrates a significant weight gain after the administration of 8.4 ml of Aloe Vera gel.

The initial weight for Group E (13.9 ml) was 86.27 ± 18.23, which increased to 131.11 ± 39.41. The t-value was -3.27, and the p-value of 0.01 again suggests a statistically significant weight increase. This shows that the higher dosage of 13.9 ml of Aloe Vera gel also leads to a significant increase in body weight.

Table 1. Mean± Standard Deviation (S.D) of Initial and Final Body Weight of Adult Wistar Administered Aloe Vera Gel

	INITIAL (Mean±S.D)	**FINAL (Mean±S.D)**	**t-Value**	**P-Value**
GROUP A (CONTROL)	93.13±14.59	163.57±33.72	-6.06	0.01
GROUP B (1.4ml)	82.53±11.91	145.91±25.58	-7.10	0.01
GROUP C (2.8ml)	83.84±20.92	180.94±30.02	-8.39	0.01
GROUP D (8.4ml)	80.02±10.72	130.74±17.47	-7.82	0.01
GROUP E (13.9ml)	86.27±18.23	131.11±39.41	-3.27	0.01

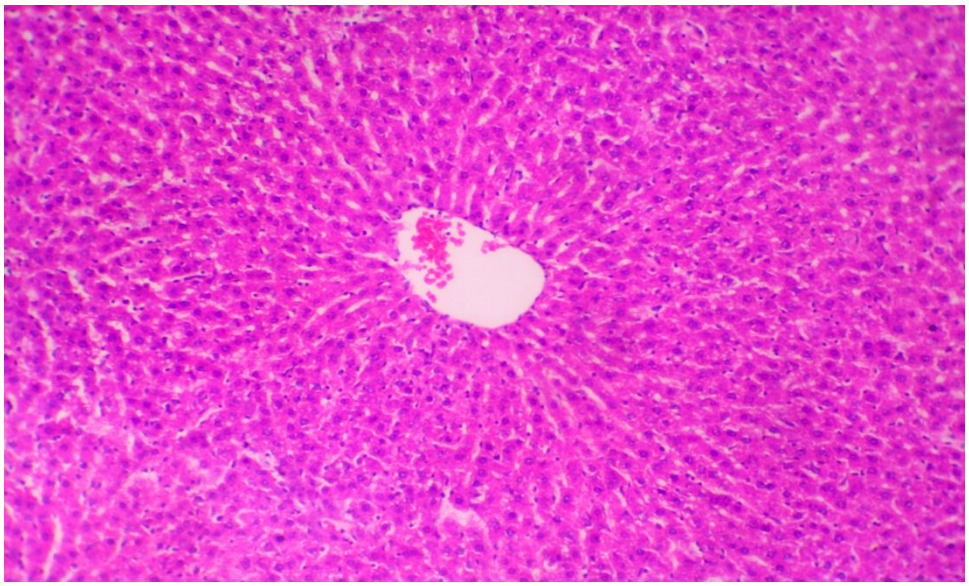

Figure 1

Plate 1a shows the Normal histological feature of the Liver of the untreated (Positive control) Albino rat with presence of Kupffer cell (A), Central vein (B) and Hepatic sinusoids (C) of the liver. HandEX100

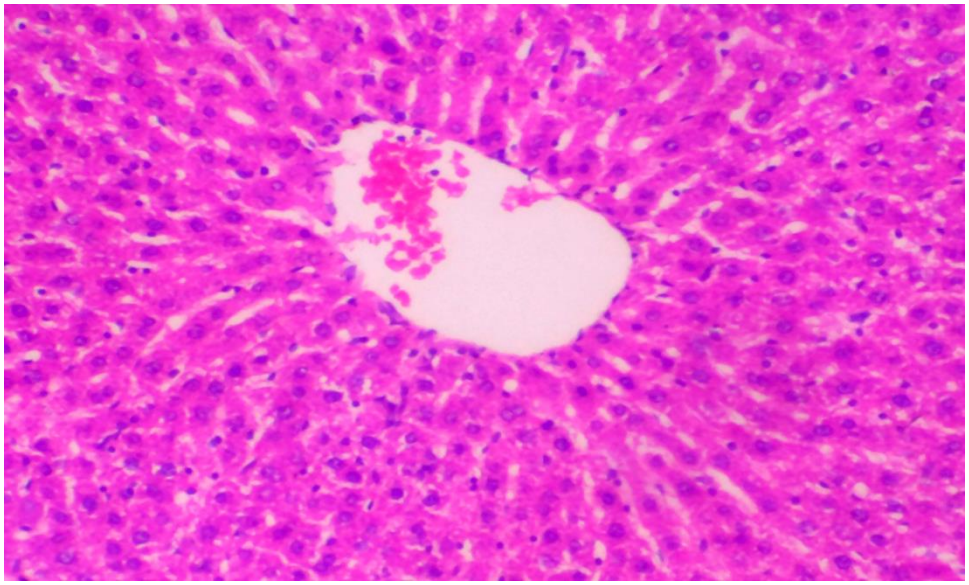

Figure 2

Plate 1b shows the Normal histological feature of the Liver of the untreated (Positive control) Albino rat with presence of Kupffer cell (A), Central vein (B) and Hepatic sinusoids (C) of the liver. HandEX400

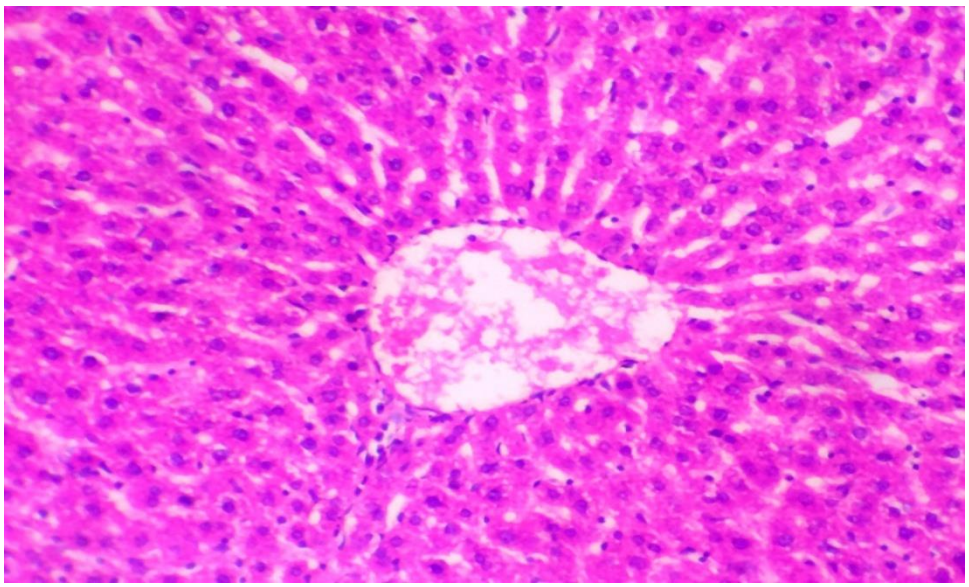

Figure 3

Plate 2a shows the Normal histological feature of the Liver of the untreated (Group B received 1.4ml of alovera gell extracts) Albino rat with presence of Kupffer cell (A), Central vein (B) and Hepatic sinusoids (C) of the liver. HandEX100

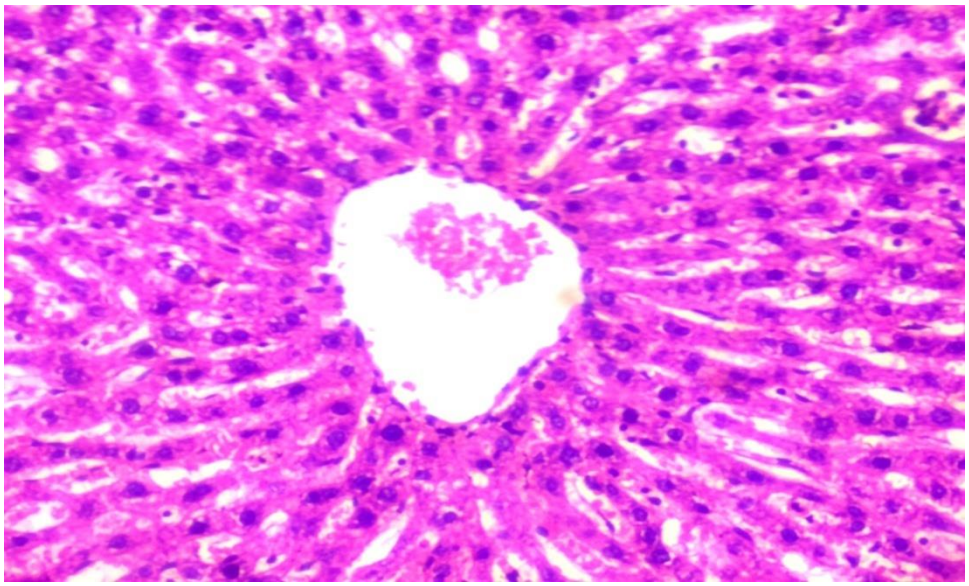

Figure 4

Plate 2b shows the Normal histological feature of the Liver of the untreated (Group B received 1.4ml of alovera gell extracts) Albino rat with presence of Kupffer cell (A), Central vein (B) and Hepatic sinusoids (C) of the liver. HandEX400

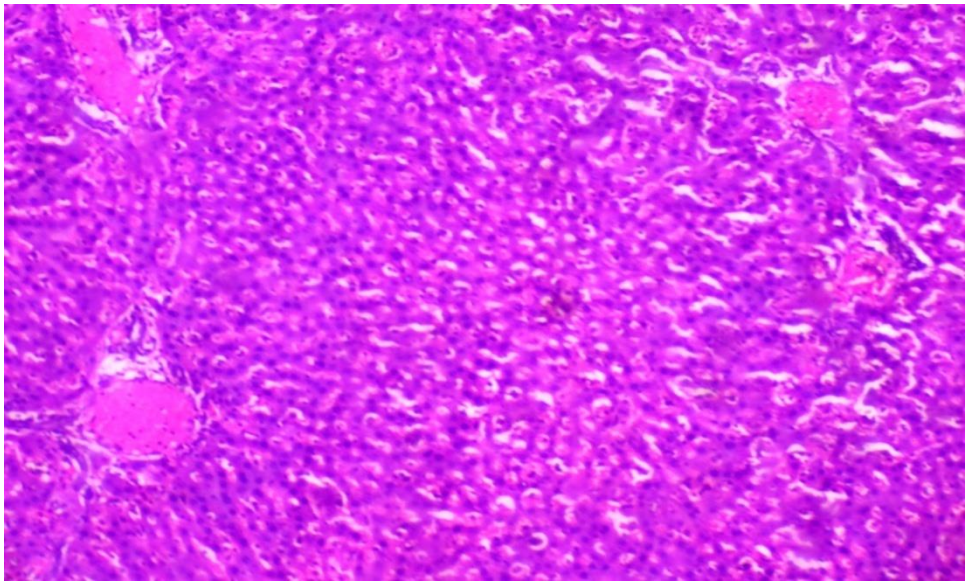

Figure 5

Plate 3a shows the histological feature of the Liver of the untreated (Group C received 2.8ml of alovera gell extracts) Albino rat with presence of Kupffer cell (A), Hepatic sinusoids (B) and Vascular congestion (C) of the liver. HandEX100

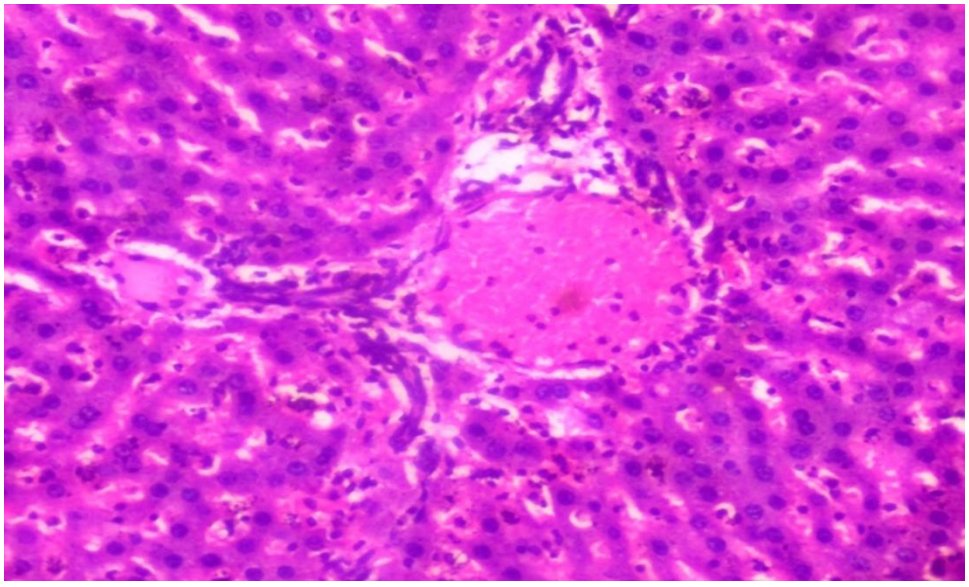

Figure 6

Plate 3b shows the histological feature of the Liver of the untreated (Group C received 2.8ml of alovera gell extracts) Albino rat with presence of Kupffer cell (C), Hepatic sinusoids (A) and Vascular congestion with presence of cellular infiltration of inflamatory cells (B) of the liver. HandEX400

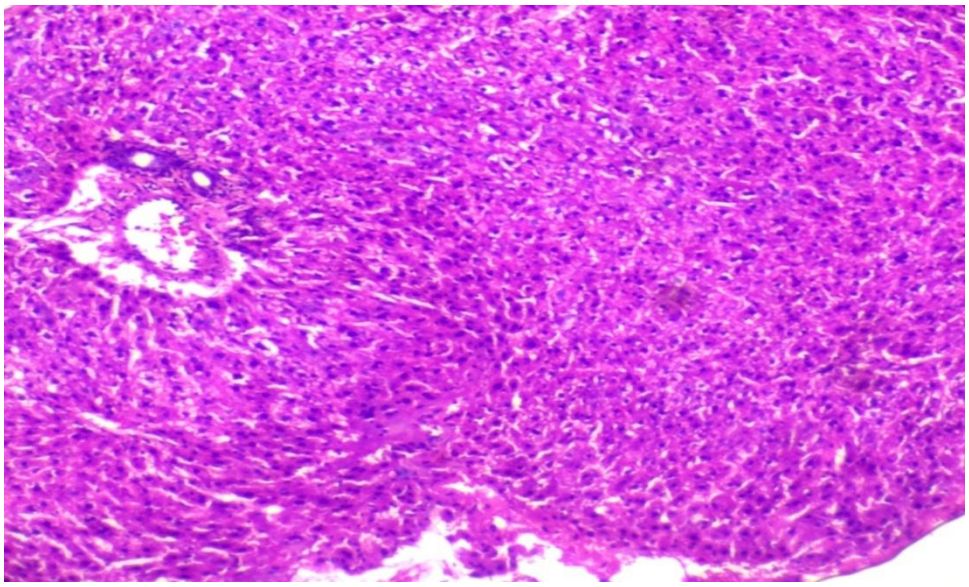

Figure 7

Plate 4a shows the histological feature of the Liver of the untreated (Group D received 8.4ml of alovera gell extracts) Albino rat with presence of Kupffer cell (A) and Diffused hepatocellular change characterized by swollen hepatocyte with pale cytoplasm infering hepatic necrosis (B) and of the liver. HandEX100

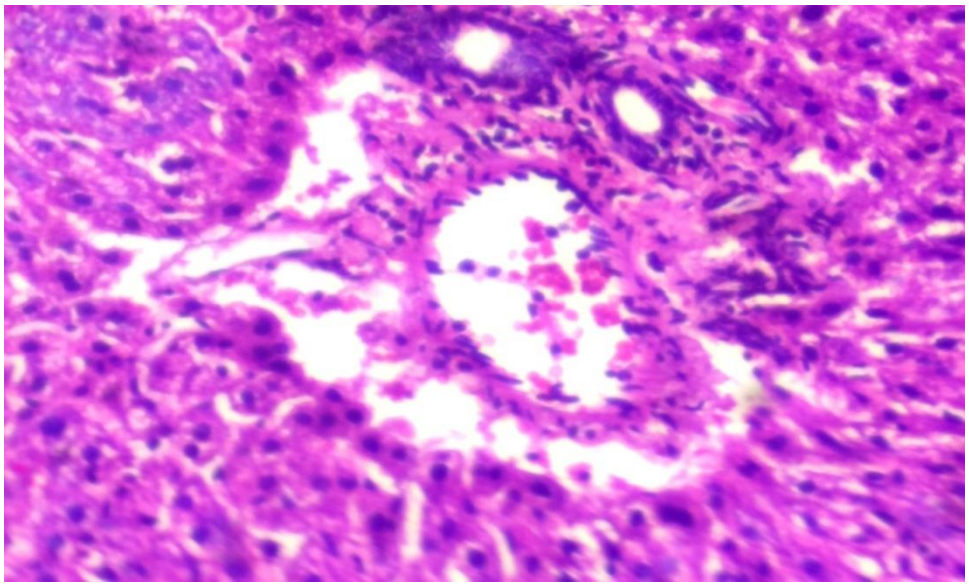

Figure 8

Plate 4b shows the histological feature of the Liver of the untreated (Group D received 8.4ml of alovera gell extracts) Albino rat with presence of Kupffer cell (A) and Diffused hepatocellular change characterized by swollen hepatocyte with pale cytoplasm infering hepatic necrosis (B) and of the liver. HandEX400

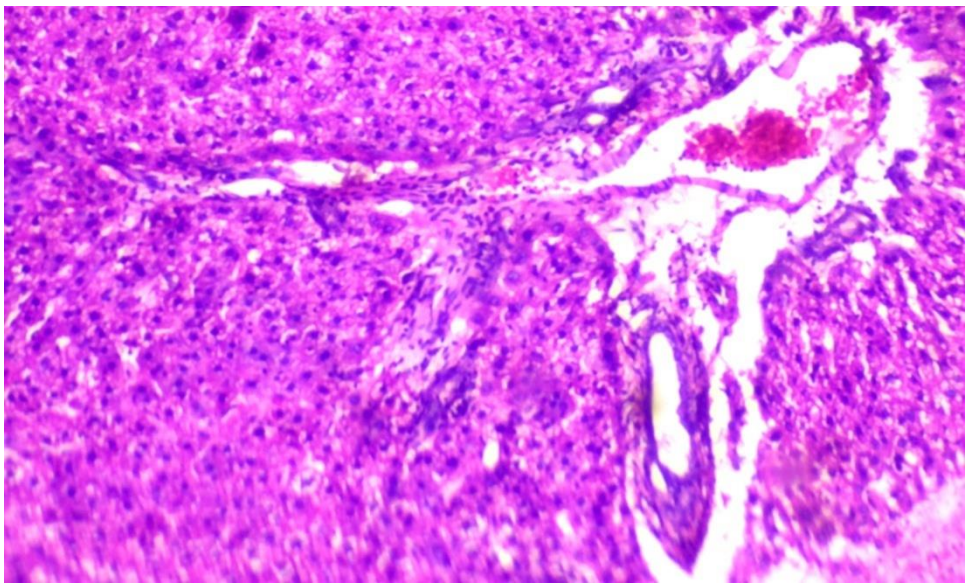

Figure 9

Plate 5a shows the histological feature of the Liver of the untreated (Group E received 13.9 ml of alovera gell extracts) Albino rat with presence of Vascular congestion (A), Kupffer cell (B) and Shrinkage of the central vein (C) of the liver. HandEX100

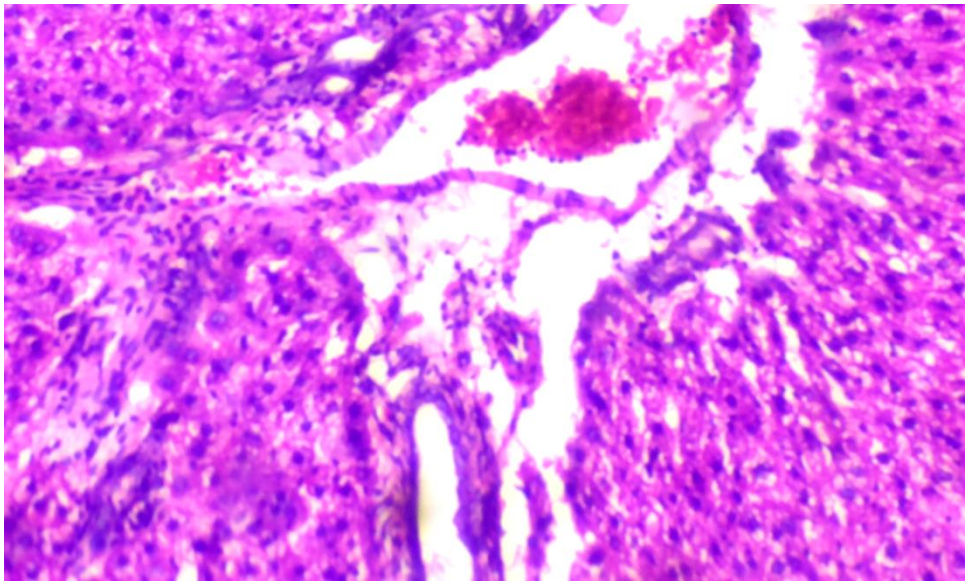

Plate 5b shows the histological feature of the Liver of the untreated (Group E received 13.9ml of aloe vera gel extracts) Albino rat with presence of Vascular congestion (A), Kupffer cell (B) and Shrinkage of the central vein (C) of the liver. HandEX400

DISCUSSION

The findings of this study demonstrate that Aloe Vera gel significantly influenced the body weight of adult Wistar rats exposed to smoke, with significant increases observed across all groups treated with varying doses of the gel. These results suggest that Aloe Vera gel may have a positive effect on body weight, which could be linked to several factors, including its anti-inflammatory properties, its ability to improve metabolic function, and the role of dosage in regulating the magnitude of its effects.

One of the possible reasons for the observed increase in body weight could be the anti-inflammatory effects of Aloe Vera gel. Aloe Vera has long been known for its potent anti-inflammatory properties, which might help mitigate the harmful effects of smoke exposure, thereby promoting recovery and weight gain. According to Shukla and Rasik (2000), Aloe Vera gel contains compounds like anthraquinones and polysaccharides that have been shown to reduce inflammation and improve tissue repair. This reduction in inflammation may contribute to better nutrient absorption and overall health, which could explain the significant increase in body weight observed in all groups, including the control group (Group A). Furthermore, Aloe Vera has been reported to enhance digestion and nutrient absorption, which could lead to a more efficient metabolism and, ultimately, weight gain (García-Pérez et al., 2018).

Another possible explanation for the weight increase could be the role of Aloe Vera in improving metabolic functions, particularly in the liver and intestines. Aloe Vera has been reported to have a hepatoprotective effect, which might aid in the efficient processing and storage of nutrients, thereby contributing to weight gain (Hsu et al., 2009). The liver's role in metabolism is crucial, and any support it receives from Aloe Vera gel could enhance the rat's

overall ability to gain weight, especially after smoke exposure, which can damage metabolic systems. In the context of smoke exposure, the healing properties of Aloe Vera could help restore the metabolic balance disrupted by the toxins in smoke, enabling the animals to recover and gain weight at a faster rate.

Interestingly, the results from Group E (13.9 ml) showed a smaller weight increase compared to the lower doses (Group C at 2.8 ml and Group D at 8.4 ml). This could suggest a dose-dependent response, where higher doses of Aloe Vera gel may have led to a regulatory effect, limiting further weight gain. In a similar study by Ghannam et al. (2015), it was found that excessive doses of Aloe Vera could lead to side effects like diarrhea or gastrointestinal disturbances, which could limit nutrient absorption and potentially affect body weight. Therefore, while moderate doses seem to have the most beneficial effects, very high doses could lead to adverse outcomes that counteract the positive effects observed at lower doses.

These findings are consistent with previous studies that demonstrate the positive effects of Aloe Vera on body weight. For example, a study by Al-Saadi and Al-Dhaheri (2019) on rats treated with Aloe Vera extract showed significant weight gain and improvement in general health parameters. Similarly, in a study by Gohar et al. (2017), rats treated with Aloe Vera exhibited increased weight due to its anti-inflammatory and metabolic-enhancing properties. However, these studies also note the potential side effects of Aloe Vera, particularly at higher doses, aligning with the more modest weight gain observed in Group E of the present study.

The histological analysis of liver tissues from experimental groups treated with Aloe vera gel extracts revealed significant variations in structural integrity, particularly at higher doses. The positive control group (Plate 1a and Plate 1b) showed normal liver architecture, including the presence of Kupffer cells, central veins, and hepatic sinusoids. Similarly, Group B, which received 1.4 ml of Aloe vera gel extract, exhibited a comparable liver structure, suggesting that a lower dose of Aloe vera gel does not induce significant hepatic alterations.

However, Group C (2.8 ml Aloe vera extract) displayed vascular congestion and cellular infiltration of inflammatory cells, indicating the onset of hepatic stress or immune response. These findings may be attributed to Aloe vera's bioactive compounds, which, at moderate doses, can trigger an inflammatory response due to their detoxifying properties (Surjushe et al., 2008). Additionally, Aloe vera has been reported to have hepatoprotective effects at controlled doses, but excessive intake may lead to oxidative stress and subsequent tissue damage (Guo & Mei, 2016).

In Group D (8.4 ml Aloe vera extract), diffuse hepatocellular changes, including swollen hepatocytes with pale cytoplasm indicative of hepatic necrosis, were observed. This suggests that higher doses of Aloe vera may have a cytotoxic effect on liver cells, potentially due to the accumulation of anthraquinones, compounds known for their hepatotoxicity at elevated concentrations (Boudreau & Beland, 2006). The presence of necrotic hepatocytes is a significant indicator of liver dysfunction, reinforcing the notion that high

concentrations of Aloe vera extracts may exceed the liver's capacity to detoxify and metabolize its bioactive components.

Group E (13.9 ml Aloe vera extract) demonstrated vascular congestion and shrinkage of the central vein, further supporting the hypothesis that excessive Aloe vera administration may contribute to circulatory disturbances within hepatic tissues. Previous studies have reported similar findings, where prolonged exposure to Aloe vera at high doses resulted in liver congestion and fibrosis in rodent models (Rajasekaran et al., 2006). This observation suggests that Aloe vera extracts, despite their beneficial properties, should be administered with caution to prevent potential hepatic complications.

These findings align with research highlighting Aloe vera's dual role in hepatic health. Studies by Chandan et al. (2007) demonstrated that Aloe vera exhibits hepatoprotective properties at low to moderate doses, aiding in liver regeneration and detoxification. However, the hepatotoxic effects observed in Groups D and E correlate with research by Yagi et al. (2009), which showed that prolonged Aloe vera administration at high doses can lead to hepatic fibrosis and necrosis, particularly in animals with pre-existing liver conditions.

CONCLUSIONS AND RECOMMENDATIONS

The study suggests that while Aloe vera gel extracts may confer hepatoprotective benefits at low doses, excessive administration poses significant risks to liver integrity. Moderate doses (such as 1.4 ml) did not result in observable liver damage, while higher doses led to vascular congestion, hepatocyte necrosis, and inflammatory infiltration. These findings underscore the importance of dose regulation when utilizing Aloe vera for therapeutic purposes, as excessive intake may lead to hepatic impairment rather than hepatoprotection.

FURTHER STUDY

Every research is subject to limitations; thus, you can explain them here and briefly provide suggestions to further investigations.

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