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Research Article

Effects of Black Seed (*Nigella Sativa*) Oil on Hepatic Function Parameters in Rats Induced with Diabetes Mellitus and Periodontitis

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ABSTRACT

This study evaluated the effect of *Nigella sativa* (NS) seed oil on hepatic function in rat model of periodontitis and diabetes mellitus. Forty-eight Wistar rats were divided into eight groups of six rats each as follows: Group I rats were fed with normal rat chow *ad libitum* without any induction and served as Control. Group II rats were fed with normal rat chow, water and administered NS oil. Group III rats were induced with diabetes without treatment. Group IV rats were treated with 1 ml/kg bwt NS oil intraperitoneally after diabetes induction (DB + NS.). Group V were induced with periodontitis without treatment. Group VI rats were treated with 1 ml/kg bwt NS oil intraperitoneally after periodontitis induction. Group VII were induced with diabetes and periodontitis without treatment and finally Group VIII rats were treated with 1 ml/kg bwt NS oil after diabetes and periodontitis induction (DB+PD+NS) intraperitoneally. The result showed that treatment with *Nigella sativa* oil significantly reduced Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) activities in Diabetes mellitus and periodontitis group compared with untreated groups. In addition, diabetes and periodontitis disrupted the normal architecture of the liver in the untreated group while the oil ameliorated these effects significantly in the treatment groups. The histology of the jaw showed varying degrees of inflammation in the untreated diabetes and periodontitis group, but these effects were reduced in the treated groups. In conclusion, black seed oil ameliorated the effect of diabetes mellitus and periodontitis in the liver of the Diabetes mellitus and periodontitis induced rats.

Keywords: *Diabetes, Periodontitis, Nigella sativa, Liver*

INTRODUCTION

Diabetes Mellitus (DM) is a chronic disease that occurs either when there is low insulin production from the pancreas or when the body cannot effectively use the insulin it produces. Insulin is the hormone that regulates blood sugar. It is a metabolic and endocrine disorder distinguished by elevated level of blood glucose (hyperglycemia) that results from inability of pancreas to produce ample amount of insulin (DM type 1) or failure of proper response to insulin (DM type 2) (Shori, 2015). Moreso, diabetes accelerates the production of reactive oxygen species (ROS)

and causes oxidative chemical modifications of lipids, DNA and proteins in various tissues (Osawa and Kato, 2005). Tissue oxidative stress and damage due to diabetes play important roles in the development of complications of diabetes. Hyperglycemic patients have 25-50% more chances to develop hepato-renal disorders, hepatic cirrhosis and carcinoma of hepatic cells due to altered serology (Hung *et al.*, 2012). Diabetes poses wide risk on the liver like non-alcohol fatty liver disease (NAFLD), severe liver scarring, liver cancer and liver failure (Keith *et al.,* 2004). Statistics has shown that 2.8 % of the world's population suffers from diabetes, with the likelihood of increasing to more than 5.4 % by 2025 (International Diabetes Federation, 2014).

Periodontitis is a chronic inflammatory disease of the periodontium that leads to tooth loss (Silva *et al.*, 2015). It is one of the most common diseases in adults. Oral bacteria and their products (e.g., lipopolysaccharide and proteases) are responsible for causing periodontitis (Madianos *et al.,* 2005). The progression of periodontal diseases also depends on the host reactions to bacterial pathogens (Page and Kornman, 2000). Moreso the damage to the tissues is irreparable. In the early stage, the problem is frequently asymptomatic; it is generally not painful and many people do not know until the condition is sufficiently progressive to result in mobility of the tooth. The pockets become deeper because fibers are further destructed in the periodontal ligament (called loss of attachment) and because the alveolar bone is reabsorbed, which occurs parallel to the loss of attachment that progresses (Rajasekar *et al.*, 2021). Gingival erythema and oedema, gingival bleeding, gingival recession, tooth mobility, tooth drifting, suppuration from periodontal pockets, and tooth loss are all symptoms of advanced periodontitis. Severe periodontitis that threatens tooth retention affects 10–15 percent of adults in the majority of populations studied (Papapanou *et al.*, 2018). There is prominent evidence that suggest that diabetes and periodontitis have a two-way relationship, with diabetes increasing the incidence of periodontitis and periodontal inflammation negatively influencing glycemic control (Preshaw *et al.,* 2012; Wahid *et al.*, 2013). Diabetes has been proven to be a significant risk factor for periodontitis (Salvi *et al.,* 2008; Chavarry *et al.,* 2009). Diabetic patients have a threefold greater incidence of periodontitis compared to non-diabetic patients (Mealey and Ocampo, 2007). The degree of glycemic control is critical in determining increased risk.

The liver plays an important role in the metabolism of all drugs; therefore, it is highly subjected to hepatotoxicity (Meyer and Kulkarni, 2001). Through hepatic metabolism, drugs are converted into active substance or products which usually have a lower pharmacologic activity and are more readily excreted than the original drug (Keith and Tolman, 2004). Untreated diabetes mellitus causes multiple histopathological changes in various organs including the liver and the incidence of diabetic hepato - dysfunction is increasing (Adewole *et al.,* 2006).

Medicinal plants have been used since ancient times as therapeutic agents for treatment of diabetes and other diseases and ailments (Belgaumi *et al.*, 2020). One of the top ranked evidence-based herbal medicines, which has been described as the "miracle herb of the century is *Nigella sativa* (Ahmad *et al.*, 2020). The medicinal assets of *N. sativa* have been established in Islamic medicine, Chinese traditional treatments, Unani, Ayurveda and other medicinal systems (Maideen, 2020; Mohebbati and Abbasnezhad, 2020). Also called black cumin, black seed is native to the south and southwest Asia, and is cultivated in several countries in the Mediterranean region, South Europe, Syria, Turkey, and Saudi Arabia. *N. sativa* is well-known to exhibit a wide spectrum of anticancer, hepatoprotective, antioxidant, antidiabetics, nephroprotective and immunomodulatory activities (Ahmad *et al.,* 2013; Hamdan *et al.*, 2019; Mekhemar *et al.*, 2020; Abd-Elkareem *et al.*, 2022). Researchers have attributed the health promoting benefits of the black seed to its active components and high nutritional content such as fixed oils, alkaloids, proteins, saponins and essential oils (Yimer *et al.*, 2019; Ahmad *et al.*, 2020). Thymoquinone is one of the pharmacologically active components that have been isolated from *N. sativa* (Ahmad *et al.*, 2020). Thymoquinone, a powerful antioxidant, reduces the development of inducible nitric oxide synthase in rat macrophages, and have been shown to have anti-diabetic properties (Alimohammadi *et al.,* 2013).

The search for natural products that offer relief from diabetes and its complications cannot be overemphasized. Though some studies have shown positive effects of *Nigella sativa* in diabetes (Asgary *et al* 2015; Hadi *et al*, in 2016; Akhtar *et al.,* 2020) and periodontitis (Al-Wafi and Shaker, 2014; Bilgic *et al.*, 2017; Kiari *et al.*, 2018; Ilangovan and Rajasekar, 2021), studies reporting its effects on hepatic function in rats induced concurrently with diabetes and periodontitis are scarce. In addition, more studies on the effects of *Nigella sativa* in diabetes are needed since it is difficult to determine effective type and dosage of *N. sativa* in diabetes management due to chemical compositions of different sources of *N. sativa*, dosage, and duration of intervention. This study is therefore designed to examine the effect of black seed oil administration on hepatic dysfunctions in diabetes mellitus and periodontitis induced rats.

MATERIALS AND METHODS

Chemicals and reagents

Nigella sativa oil manufactured by Hemani international KEPZ Karachi Pakistan was purchased. ALT, ALP and AST kits were products of Randox Laboratories, UK. Streptozotocin, Heparin injection, Saline, distilled water, urethane, chloralhydrate, formaldehyde, formic acid and all other chemicals and regents used were of analytical grade and were obtained from B.D.H, Poole England.

Animals

The study was carried out with 48 adult-male Wistar rats weighing140–160 g. For adaptation, the animals were kept

in the animal house of the College of Medicine, University of Lagos, Lagos State for 2 weeks before the start of the experiment. The Wistar rats were kept in wire mesh cages and were allowed free access to water and food pellets (grower's mash) at a room temperature of about 29°C ± 2°C throughout the period of this experiment. They had a 12-hour light and 12-hour dark cycle. All applicable international, national, and/or institutional guidelines for the care and use of animals were followed (Olfert *et al.*, 1993).

Animals grouping and treatments

A total of forty-eight (48) Wistar rats were divided into 8 groups of 6 rats each as follows: Group I rats were fed normal rat chow *ad libitum* without any induction and served as Control. Group II rats were fed normal rat chow with water and administered NS oil. Group III rats were induced with diabetes without treatment. Group IV rats were treated with 1 ml/kg body weight NS oil (Orororo *et al.*, 2023) intraperitoneally after diabetes induction (DB + NS). Group V were induced with periodontitis without treatment. Group VI rats were treated with 1 ml/kg bwt NS oil intraperitoneally after periodontitis induction. Group VII were induced with diabetes and periodontitis without treatment and finally Group VIII rats were treated with 1 ml/kg bwt NS oil after diabetes and periodontitis induction (DB + PD+NS1) intraperitoneally. This arrangement is illustrated in Table 1.

Table 1. Animal Groups and Numbers

Groups	Name	Induction	Treatment	Numbers
1	Normal Control	Normal chow + water	No treatment	6
2	Treatment	Normal chow + water	Oral *N. sativa* oil admin.	6
3	Diabetic	Injection of STZ	No treatment	6
4	Diabetic+ Treatment	Injection of STZ	Oral *N. sativa* oil admin.	6
5	Periodontitis	Ligature-induced	No treatment	6
6	Periodontitis+ Treatment	Ligature-induced	Oral *N. sativa* oil admin.	6
7	Diabetes+ Periodontitis	Injection of STZ and ligature induction of periodontitis	No treatment	6
8	Diabetes+ Periodontitis + Treatment	Injection of STZ and ligature induction of periodontitis	Oral *N. sativa* oil admin.	6

Induction of experimental diabetes

Experimental animals were fasted overnight before induction of diabetes with Streptozotocin. The rats were given single intraperitoneal injection of a freshly buffered (0.1 m citrate, pH, 4.5) solution of streptozotocin (STZ) at a dosage of 50 mg/kg body weight. After 72 h of STZ administration, the tail vein blood was collected to determine blood glucose level with the aid of a very sensitive glucometer (Orororo *et al.*, 2023).

Induction of periodontitis

Silk suture (3/0) was inserted subgingivally on the incisor of all rats for ligature-induced periodontitis (Hatipoglu *et al.,* 2015), under general anaesthesia (Chlorahydrate 30mg/kg of body weight). The ligature acted as a gingival irritant for twenty-one (21) days and promoted the accumulation of plaque and subsequently development of periodontal disease. After placing the ligatures, the animals were observed for 21 days, and daily ligature checks were performed.

Administration of nigella sativa

The oil was administered at 1ml/kg body weight of experimental animals intraperitoneally for 21 days in the treatment groups (Mohamed *et al.*, 2010). Animals were fasted for one day prior to dosing.

Blood sample collection

Each group of animals were sacrificed after completion of the expected exposure and treatment periods. After overnight fasting, rats were sacrificed following euthanisation by cervical dislocation (Morakinyo *et al.*, 2014). Blood was collected via cardiac puncture into plain bottles and centrifuged immediately at 3000 rpm for 10 min to obtain serum for estimation of liver function tests.

Serum analysis

Liver activity parameters (Aspartate aminotransferase, Alanine aminotransferase, and Alkaline phosphatase) were measured using commercially available diagnostic kits. Aspartate and alanine aminotransferases were measured by the method of Reitman and Frankel (1957), where the transfer of the amino group from aspartate or alanine formed oxalacetate or pyruvate, respectively and the developed colour was measured at 520 nm.

Histological evaluation of the liver

Following fixation, tissues were cut into thin slices ($1cm^3$) with the help of a clean sharp knife. Tissues were processed by paraffin technique, sectioned at 5-7 µm and subjected to Hematoxylin and Eosin (H&E) staining procedure (Bancroft *et al.*, 2013). Slides were examined under microscope (Olympus Cx23 Binocular) at 100X to measure the thickness (µm) of endometrium and myometrium using automated image analysis system, image. Vacuolar and degenerative changes in the liver, radial arrangement of hepatocytes

around central vein and necrosis of hepatocytes were also observed.

Histological evaluation of the jaw

After the rats were sacrificed, the lower jaws were extracted and fixed for 72 hours in 10% formalin. The jaw was rinsed with normal saline then fixed in 10% formal-saline, dehydrated in graded alcohol, and embedded in paraffin. The jaw tissues were cut into 5µm thick sections and stained with haematoxylin-eosin for photo microscopic assessment (Bancroft *et al.*, 2013).

Statistical analysis

The data were analyzed using ANOVA and SNK posthoc test. Significance level set at $p<0.05$. Data analysis was carried using Graphpad Prism 9.

RESULTS AND DISCUSSION

Effects of nigella sativa oil on aspartate aminotransferase (AST) in rats induced with diabetes mellitus and periodontitis

Figure 1 shows a significant ($p<0.05$) elevation of aspartate aminotransferase in the untreated Diabetes mellitus and Diabetes mellitus + periodontitis (DM +PD) groups, respectively, compared with the Control group. In the Periodontitis (PD) group, no significant difference was seen compared with the Control. Moreso, aspartate aminotransferase was mildly reduced in the Diabetes mellitus and Periodontitis groups administered *Nigella sativa* oil compared with the untreated Diabetes mellitus and Periodontitis groups.

Effects of nigella sativa oil on alanine aminotransferase (AST) in rats induced with diabetes mellitus and periodontitis

The bar chart below (Figure 2) shows a significant elevation ($p<0.01$) of Alanine aminotransferase in the untreated Diabetes mellitus and Diabetes mellitus + periodontitis groups, respectively, compared with the untreated Control group. In the Periodontitis (PD) group, no significant difference was seen compared with the untreated Control. Moreso, Alanine aminotransferase was significantly reduced in Diabetes mellitus and Periodontitis groups administered intraperitoneal *Nigella sativa* oil compared with the untreated Diabetes mellitus and Periodontitis groups.

Effects of nigella sativa oil on alkaline phosphatase in rats induced with diabetes mellitus and periodontitis

The effect of *Nigella Sativa* Oil on alkaline phosphatase in experimental rats were reduced, and the results are presented in Figure 3. There was a non-significant increase in ALP in rats induced with DM alone relative to the untreated Control group. Also, there was no significant increase in ALP in PD alone induced rats compared to the untreated Control group. Moreso, significant elevation was observed in rats with Periodontitis + Diabetes mellitus compared with untreated Control. The administration *Nigella Sativa* Oil after DM and PD induction caused decreased ALP compared to the untreated Control group. There was also no significant reduction in ALP level in rats administered *Nigella Sativa* Oil after DM induction when compared to the untreated Control group.

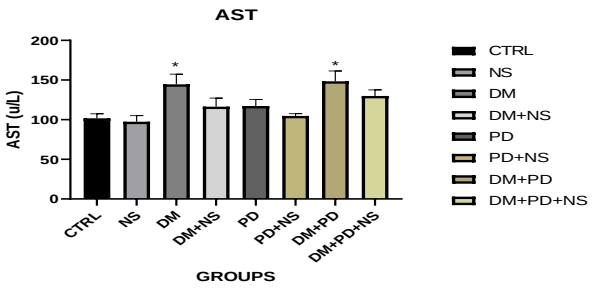


Figure 1. Effects of NS Oil on Aspartate Aminotransferase (AST) Across the Groups.

Values are Mean ± SEM (n=5). *=Significantly higher ($p<0.05$) in DM group vs Control. **=Significantly higher ($p<0.01$) in DM+PD group vs Control. CTRL=Control, NS=*Nigella sativa*, DM= Diabetes mellitus, DM+NS=Diabetes mellitus+Nigella sativa, PD=Periodontitis, PD+NS= Periodontitis+ Nigella sativa, DM+PD= Diabetes mellitus+ Periodontitis, DM+PD+NS= Diabetes mellitus+ Periodontitis+Nigella sativa.

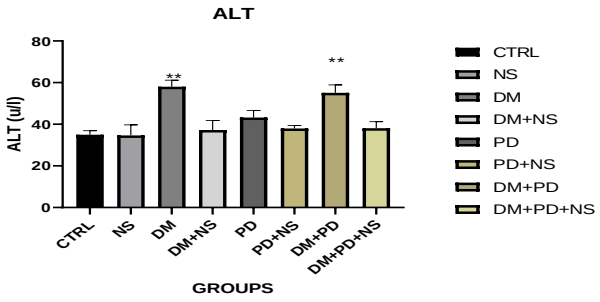


Figure 2. Effects of NS Oil on Alkaline Phosphatase Across the Groups

CTRL=Control, NS=Nigella sativa,DM= Diabetes mellitus, DM+NS=Diabetes mellitus+Nigella sativa, PD=Periodontitis, PD+NS= Periodontitis+ Nigella sativa, DM+PD= Diabetes mellitus+ Periodontitis, DM+PD+NS= Diabetes mellitus+ Periodontitis+Nigella sativa

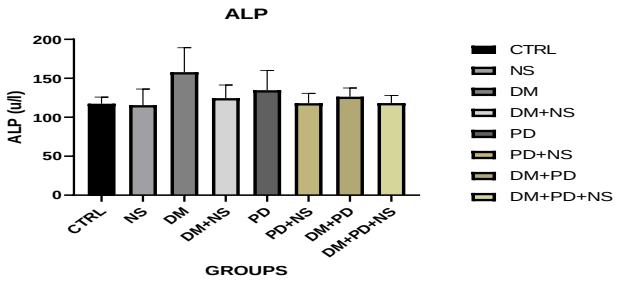


Figure 3. Effects of NS Oil on Alkaline Phosphatase Across the Groups.

CTRL=Control, NS=Nigella sativa,DM= Diabetes mellitus, DM+NS=Diabetes mellitus+Nigella sativa, PD=Periodontitis, PD+NS= Periodontitis+ Nigella sativa, DM+PD= Diabetes mellitus+ Periodontitis, DM+PD+NS= Diabetes mellitus+ Periodontitis+Nigella sativ

Effects of nigella sativa oil on histology of the liver in rats induced with diabetes mellitus and periodontitis

The effects of *Nigella sativa* Oil on histology of the liver in rats induced with Diabetes mellitus and Periodontitis are shown in Plates 1- 8. Normal architecture consisting of central vein (CV), portal triad (PT) was seen in liver section of rat from control group and *Nigella sativa* treatment groups, whereas photomicrograph of liver section of rat from Diabetic and periodontitis groups showed peliosis hepatis, lobular inflammation, sinusoidal dilatation, portal tract inflammation with interface hepatitis. Administration of the oil to induced rats had restorative effects.

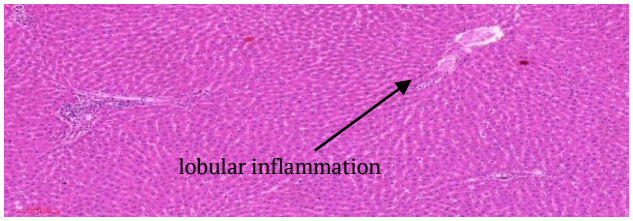
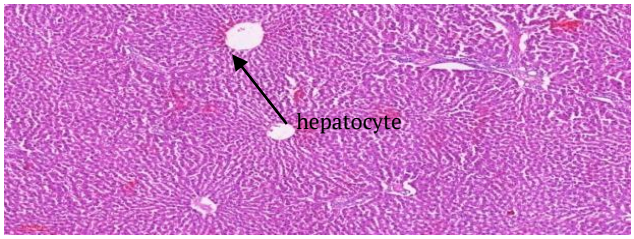
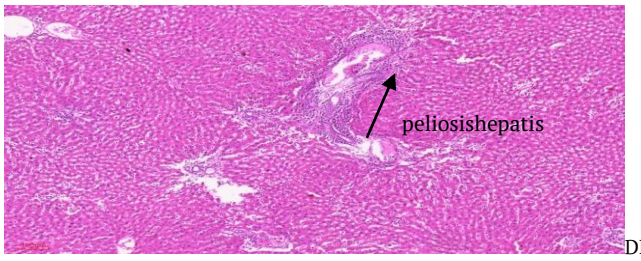


Plate 1. Photomicrograph of liver section of rat from control group displaying normal architecture, central vein (CV), portal triad (PT) and few lobular inflammation (H&E, X100).



NS

Plate 2. Photomicrograph of liver section of rat from *Nigella sativa* treatment group displaying central vein (CV), sinusoid (Si), and hepatocyte (H&E, X100).



DB

Plate 3. Photomicrograph of liver section of rat from diabetic group showing peliosis hepatis, lobular inflammation, sinusoidal dilatation, portal tract inflammation with interface hepatitis (H&E, X100).

Plate 4. Photomicrograph of liver section of rat from diabetes + treatment group showing significantly reduced portal tract and lobular inflammation, no interface with hepatitis (H&E, X100).

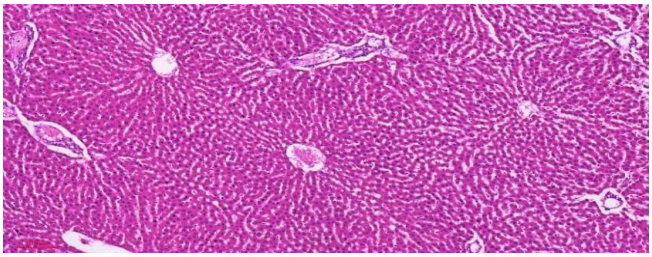


Plate 5. Photomicrograph of liver section of rat from periodontitis group showing minimal portal tract inflammation near normal (H&E, x 100).

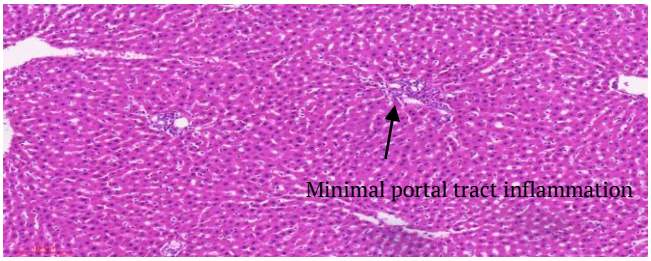


Plate 6: Photomicrograph of liver section of rat from periodontitis and treatment group showing minimal portal tract inflammation (H & E, x 100).

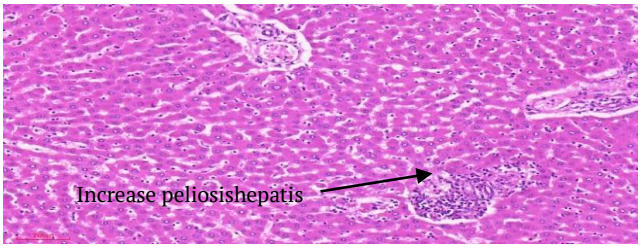


Plate 7. Photomicrograph of liver section of rat from Periodontitis and diabetes induced group displaying increased peliosis hepatis, lobular inflammation, sinusoidal dilatation, portal tract inflammation with interface hepatitis (H&E, X100).

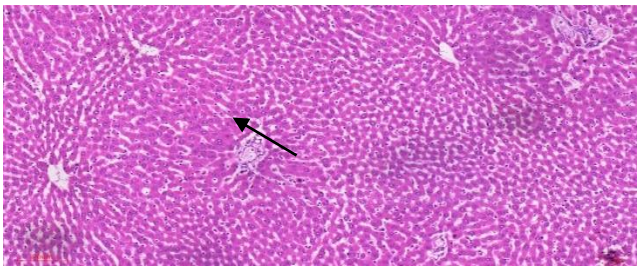


Plate 8. Photomicrograph of liver section of rat from Periodontitis + diabetes and treatment group showing normal architecture, central vein (CV), portal triad (PT) and few lobular inflammation(H&E, X100).

Effects of *nigella sativa* oil on histology of the jaw in rats induced with diabetes mellitus and periodontitis

The Effects of *Nigella sativa* Oil on histology of the Jaw in rats induced with Diabetes mellitus and Periodontitis is shown in Plates 9-16. Few periodontal neutrophil aggregates and chronic inflammation were seen in Group One while the jaw section of rat from *Nigella sativa* group showed no periodontal neutrophil aggregate. Conversely periodontal inflammation, neutrophil, lymphocyte and plasma cells were observed in the jaw section of rat from diabetic and periodontitis groups separately and when induced together.

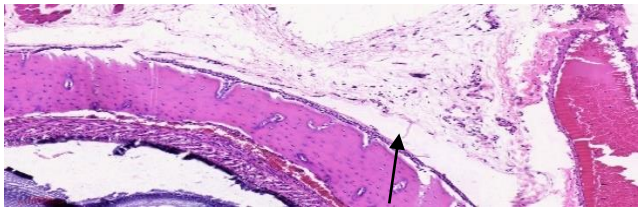


Plate 9. Photomicrograph of jaw section of rat from the control group showing few periodontal neutrophil aggregates and chronic inflammation.
(H&E x100)

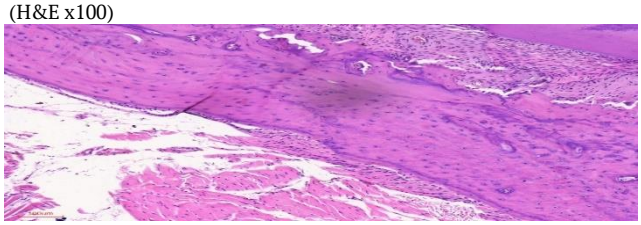


Plate 10. Photomicrograph of jaw section of rat from *Nigella sativa* group showing no periodontal neutrophil aggregate. (H&E x100).

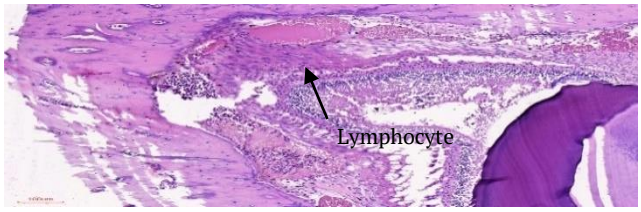


Plate 11. Photomicrograph of jaw section of rat from diabetic group showing periodontal inflammation, neutrophil, lymphocyte and plasma cells (H&E x100).

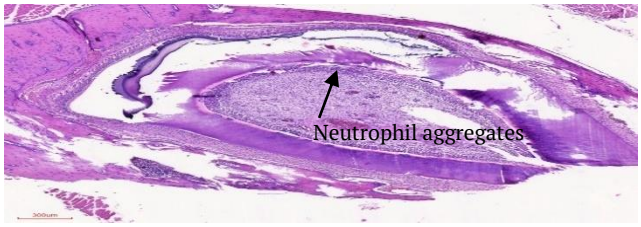


Plate 12. Photomicrograph of jaw section of rat from diabetes + treatment group few periodontal neutrophil aggregates and chronic inflammation (H&E x100).

Plate 13. Photomicrograph of jaw section of rat from periodontitis group showing periodontal inflammation mainly neutrophils, epithelial and soft tissue infiltration by inflammatory cells
(H & E x 100)

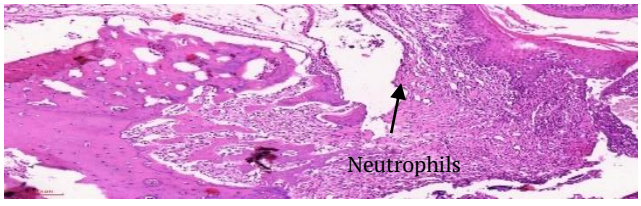


Plate 14. Photomicrograph of jaw section of rat from periodontitis + *Nigella sativa* group showing milder Periodontal inflammation mainly neutrophils, epithelial and soft tissue infiltration by inflammatory cells
(H & E x 100).

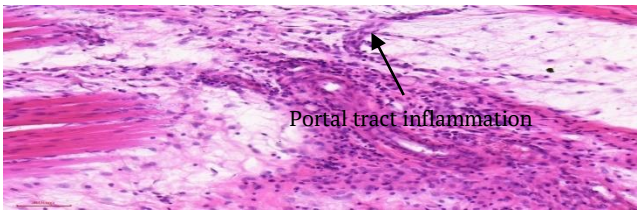


Plate 15: Photomicrograph of jaw section of rat from periodontitis and diabetes group showing periodontal inflammation mainly neutrophils, epithelial and soft tissue infiltration by inflammatory cells
(H & E x 100).

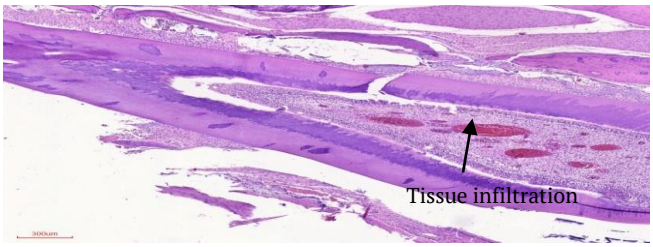


Plate 16: Photomicrograph of jaw section of rat from periodontitis + diabetes + *Nigella sativa* group showing significant reduced periodontal inflammation and tissue infiltration by inflammatory cells.
(H&E x100)

Treatment of rats induced with diabetes and periodontitis with NS oil caused significant reduction in periodontal inflammation and tissue infiltration by inflammatory cells.

Discussion

In this study, rats with diabetes mellitus and periodontitis were used as test subjects, and the effects of black seed (*Nigella sativa*) oil were assessed on liver function indicators. When compared to the control, the induction of diabetes and periodontitis markedly changed the serum levels of AST and ALT activity. The untreated Diabetes mellitus group had significantly higher levels of Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) activity in comparison to the control group. This result is consistent with previous studies (Hamad *et al.*, 2011; Usman *et al.*, 2017). A slight increase in this enzyme was also observed in the group with periodontitis, which is consistent with the findings of the Carvalho *et al.* (2016). Whereas ALT is a specific sign for hepatic parenchymal injury, AST is a generic marker for hepatic injury. Both of them are employed in the assessment of liver problems (Sepodes *et al.*, 2004; Bi *et al.*, 2008). Additionally, the untreated DM group showed a little increase in Alkaline phosphatase activity when compared to the Control. Kemink *et al.* (2000) found a comparable outcome. Increased levels of the membrane-bound glycoprotein ALP have been linked to hepatocyte membrane dysfunctions, such as impaired membrane permeability and metabolite transport (Drotman and Lawhorn, 1978; Mehana *et al.*, 2012). ALP, which is mostly found in the liver's bile duct, is thought to be a sign of cholestasis, hepatic function, and biliary function (Whitehead *et al.*, 1999, Elizabeth and Harris, 2005). Furthermore, it is possible to link the increase

in small intestine alkaline phosphatase activity in diabetic rats to an increase in blood ALP activity (Unakami *et al.*, 1990; Mansour *et al.*, 2002).

According to Bujanda *et al.* (2008), elevated liver enzyme levels are indicators of the detrimental effects of diabetes mellitus and periodontitis on the cellular makeup of hepatocytes, which release the enzymes aspartate aminotransferase, alkaline phosphatase, and alanine aminotransferase into the bloodstream. According to Ohaeri (2001), rats' livers necrosed after they were made diabetic. According to Navarro *et al.* (1993), this suggests that the rises in AST and ALT activity could be the consequence of these aminotransferase enzymes leaking into the bloodstream from the liver's cytosol. The findings of this investigation are consistent with those of other researchers (Zafar *et al.*, 2009; Najla *et al.*, 2012; Soliman, 2013; Omonkhua *et al.*, 2014) who reported similar elevations in liver enzyme activity after the creation of diabetes. Since the liver serves as the primary processing organ for fuels whose metabolism has been significantly affected by diabetes, chronic uncontrolled diabetes has been shown to cause injury and destruction to the liver (Eluehike *et al.*, 2022).

Hyperglycemia is the result of systemic and metabolic abnormalities brought on by diabetes. It causes the body to produce free radicals that weaken the antioxidant system and interfere with normal lipid, protein, and glucose metabolism (Alimohammadi *et al.*, 2013). Conversely, periodontitis is a microbiome-associated dental illness broug.

CONCLUSION

The biochemical and micrograph findings suggest that *Nigella sativa* oil (probably due to its bioactive contents) have therapeutic effects and can protect against hepatic dysfunctions in diabetes mellitus and periodontitis induced rats.

AUTHORS' CONTRIBUTIONS

Authors WAA, AAB, KB and UAO designed the experiment. UAO, OIE and CCO carried out the laboratory work. Authors OCO, EPA, OBO, EOO and EOE wrote and edited the manuscript. All the authors read and approved the final version for publication.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest

REFERENCES

Abd-Elkareem, M., Soliman, M, Abd El-Rahman, M. A. M. and Abou Khalil NS (2022) Effect of *Nigella sativa* l. Seed on the kidney of monosodium glutamate challenged rats. *Frontiers in. Pharmacology,* 13:789988. doi: 10.3389/fphar.2022.789988

Adewole, S. O., Caxton-Martins, E. A. and Ojewole, J. A. O. (2006). Histochemical and biochemical effects of melatonin on pancreatic-cells on streptozocin –treated diabetic rats. *Pharmacology-Online*, 2: 1-21

Ahmad, A., Husain, A., Mujeeb, M., Khan, S.A., Najmi, A.K., Siddique, N.A., Damanhouri, Z.A. and Anwar, F (2013). A review on therapeutic potential of *Nigella sativa*: A miracle herb. *Asian Pacific Journal of Tropical Biomedicine,* 3, 337–352.

Ahmad, M.F., Ahmad, F.A., Ashraf, S.A., Saad, H.H., Wahab, S., Khan, M.I., Ali, M., Mohan, S., Hakeem, K.R. and Athar, M.T. (2020). An updated knowledge of black seed (*Nigella sativa Linn*): Review of phytochemical constituents and pharmacological properties. *Journal of Herb Medicine,* 2: 100-110.

Akhtar, M. T., Qadir, R. Bukhari, I., Ashraf1, R. A, Zahoor, Z. M., Murtaza, M. A., Siddique, F., Shah, S. N. H. and Saadia, M. (2020). Antidiabetic potential of *Nigella sativa* L seed oil in alloxan-induced diabetic rabbits. *Tropical Journal of Pharmaceutical Research*; 19 (2): 283-289.

Al-Bayaty F., Kamaruddin A., Ismail M. and Abdulla M. (2013). Formulation and Evaluation of a New Biodegradable Periodontal Chip Containing Thymoquinone in a Chitosan Base for the Management of Chronic Periodontitis. *Journal of Nanomaterials*, 39(7):308-320.

Alimohammadi, S, Hobbenaghi, R. and Javanbakht, J. (2013). Protective and antidiabetic effect of extract from *Nigella sativa* on blood glucose concentrations against streptoztocin (STZ)-induced diabetic rats: an experimental study with histopathological evaluation. *Diagnostic Pathology,* 8:137-143.

Al-Logmani, A. and Zari, T. (2011). Long-term effects of Nigella sativa L. oil on some physiological parameters in normal and streptozotocin-induced diabetic rats. *Journal of Diabetes Mellitus*, 1: 46-53.

Al-Wafi, H. and Shaker A., (2014). Benefits of thymoquinone, a *Nigella Sativa* extract in preventing dental caries initiation and improving gingival health. *ProQuest LLC* 2014: 72.

Asgary S., Sahebkar A. and Goli-Malekabadi N. (2015). Ameliorative effects of *Nigella sativa* on dyslipidemia. *Journal of Endocrinology Investigation,* 38(10):1039-46.

Bancroft, S. L., Bancroft, J. and Gamble, M., (2013). Tissue processing. Bancroft's Theory and Practice of Histological Techniques. 7nd ed. Netherlands, Amsterdam*: Elsevier Health Sciences,* 105-123.

Baragob, A. E. A, (2015). Composition and hypoglycemic effect of CM in streptozotocin- diabetic rats. *Biochemistry and Biotechnology Research;* 3:38-42.

Belgaumi, U.I., Patil, S. and Gandhi, J. M. (2020). The many therapeutic applications of *Nigella sativa*- a review of literature. *Journal of Evolution Medicine and Dental Science,* 9(30):2151-2157.

Bilgic, S., Tastemir, D. K., Azirak, S., Guvenc A. N., Kocaman, N. and Ozer M. K. (2017). The protective effect of thymoquinone over olanzapine-induced side effects in liver, and metabolic side effects. *Bratisl Lek Listy,* 118(10): 618-625.

Bujanda, L., Hijona, E. and Larzabal, M. (2008). Resveratol inhibits nonalcoholic fatty liver disease in rats. *BMC Gastroentrology,* 8:40-8.

Busari, A. A., Orororo, O. C., Opute, U. A., Efejene, I. O., Badmos, K., Obadiah, C. C., Akinshipo, W. A. and Egbune, E. O. (2023). Effects of black seed oil on oxidative stress parameters and gingival expression of inducible nitric oxide synthase in diabetes and periodontitis-induced rats. *African Scientist*, 24(3): 376-386.

Carvalho, J.S., Di Lenardo, D., Alves, E.H.P., Carvalho, A.S., Silva, F.R.P., França, L.F.C., Nóbrega Neto, A.P.R., Vasconcelos, A.C.C.G., Souza, L.K.M., Barbosa, A.L.R., Medeiros J.V.R., Oliveira, J.S. and Vasconcelos, D.F.P. (2016) Steatosis caused by experimental periodontitis is reversible after removal of ligature in rats. *Journal of Periodontal Research*; 8(3):13-25.

Chavarry, N.G.M., Vettore, M.V., Sansone C. and Sheiham, A (2009). The relationship between diabetes mellitus and destructive periodontal disease: A meta-analysis. *Oral Health and Preventive Dental*, 7:107–127

Cheng, D., Liang B. and Li, Y. (2013). Antihyperglycemic effect of *Ginkgo biloba* extract in streptozotocin-induced diabetes in rats. *Biomedical Research International*, 6:1-7.

Ekakitie, L. I., Okpoghono, J., Orororo O. C. and Ekakitie, O. A. (2021) Ameliorative prowess of bee honey in the tissues of rats administered aluminium nitrate. *Scientific African;* 12 (2021) e00782:1-9

Elizabeth, H. and Harris, M. D. (2005). Elevated liver function tests in type 2 diabetes. *Clinical Diabetes*, 23: 115-11

Hadi, S., Mirmiran, P. and Hadi, V, (2016). Effects of *Nigella sativa* on cardiovascular risk factors. *Journal of Mazandaran University of Medical Science,* 25(134):430-442.

Hamad, E. M., Abdel-Rahim, E. A. and Romeih, E. A. (2011). Beneficial effect of CM on liver and kidneys function in diabetic Sprague-Dawley rats. *International Journal of Dairy Science,* 6:190-7.

Hamdan, A. Idrus R. H. and Mokhtar M. H. (2019). Effects of *Nigella sativa* on type-2 diabetes mellitus: A systematic review. *International Journal of Environmental Research and Public Health,* 16: 49-61.

Hatipoglu, M., Alptekin, N.Ö., Avunduk, M.C. (2016). Effects of alpha-tocopherol on gingival expression of inducible nitric oxide synthase in the rats with experimental periodontitis and diabetes. *Nigerian Journal of Clinical Practice*, 19:480-5. 2016.

Hung, H. Y., Qian, K., Morris-Natschke, S.L., Hsu, C.S. and Lee, K. H. (2012). Recent discovery of plant-derived anti-diabetic natural products. *National Production Report*; 29(5): 580-606.

Ilangovan, S. and Rajasekar, A. (2021). Evaluation of Efficacy of 20% *Nigella sativa* on Gingival Health. *Journal of Pharmaceutical Research International;* 33(61A): 354-363,

International Diabetes Federation (IDF) (2017). Diabetes atlas.8th ed. Brussels: *International Diabetes Federation*; 2017.

Kassab, R.B. and El-Hennamy, R.E. (2017). The role of thymoquinone as a potent antioxidant in ameliorating the neurotoxic effect of sodium arsenate in female rat. *Egyptian Journal of Basic and Applied Sciences,* 4, 160–167.

Keith, G. and Tolman, M.D. (2004). Hepatotoxicity of non-narcotic analgesics. *The American Journal of Medicine*; 105(1): S213-S219.

Keith G., Tolman M.D., Fonseca, A.V., Meng, H. and Dalpiaz, A. (2004). Narrative review hepatobiliary disease in type 2 diabetes mellitus. *Animals of internal Medicine*; 12, 141, 946 – 952.

Kemink, S.A.G., Hermus A.R.M.M., Swinkels L.M.J.W, Lutterman J.A. and Smals, A.G.H (2000). Osteopenia in insulin-dependent diabetes mellitus; prevalence and aspects of pathophysiology. *Journal of Endocrinology Investigation,* 23: 295-303.

Kiari, F. Z., Meddah, B. and Meddah, A. T. T. (2018) In vitro study on the activity of essential oil and methanolic extract from Algerian *Nigella sativa* L. Seeds on the growth kinetics of micro-organisms isolated from the buccal cavities of periodontal patients. *Saudi Dental Journal*; 30, 312–323.

Madianos, P. N., Bobetsis, Y. A. and Kinane, D. F. (2005). Generation of inflammatory stimuli: How bacteria set up inflammatory responses in the gingiva. *Journal of Clinical Periodontology,* 32:57-71.

Mahmoud, Y.K. and Abdelrazek, H.M.A. (2019). Cancer: Thymoquinone antioxidant/pro-oxidant effect as potential anticancer remedy. *Biomedical Pharmacotherapy*, 115, 108783.

Maideen, N.M.P. (2020). Prophetic Medicine-Nigella Sativa (Black cumin seeds) - Potential herb for COVID-19? *Journal of Pharmacopuncture,* 23, 62–70.

Mansour, M.A., Nagi, M.N. and El-Khatib, A.S, (2002). Effects of thymoquinone on antioxidant enzyme activities, lipid peroxidation and DT-diaphorase in different tissues of mice: A possible mechanism of action. *Cell Biochemical Functions,* 20:143–151.

Marchesan, J.T., Girnary, M.S., Moss, K.; Monaghan, E.T., Egnatz, G.J., Jiao, Y.; Zhang, S.;,Beck, J. and Swanson, K.V. (2020). Role of inflammasomes in the pathogenesis of periodontal disease and therapeutics. *Periodontology*, 82, 93–114.

Mazumdar, M., Chatterjee, A., Majumdar, S., Chandrika, M. and Patki, P. S. (2013). Evaluation of the safety and efficacy of complete care herbal toothpaste in controlling dental plaque, gingival bleeding and periodontal diseases. *Journal of Homeop Ayurv Medicine,* 2: 124.

Mealey, B. L. and Ocampo, G. L. (2007). Diabetes mellitus and periodontal disease. *Periodontology*; 44:127–153.

Mehana, E. E., Meki, A. R. and Fazili, K. M. (2012). Ameliorated effects of green tea extract on lead induced liver toxicity in rats. *Expert Toxicology and Pathology*, 64(4): 291-295

Mekhemar M., Hassan Y. and Dörfer C. (2020*). Nigella sativa* and *thymoquinone:* A natural blessing for periodontal therapy. *Antioxidants*, 9:12-30

Meyer S.A. and Kulkarni A.P. (2001). Hepatotoxicity. In: Introduction to biochemical toxicology. 3rd Edn. Newyok: John Wiley and Sons, p487.

Mohamed, E. (2010). Radioprotective effects of black seed (*Nigella sativa*) oil against hemopoietic damage and immunosuppression in gamma-irradiated rats. *Journal of Pathology,* 200:647–655.

Mohebbati, R. and Abbasnezhad, A. (2020). Effects of *Nigella sativa* on endothelial dysfunction in diabetes mellitus: A review. *Journal of Ethnopharmacology*, 25 (2):112-120.

Morakinyo, A. O., Adekunbi, D. A., Dada, K. A. and Adegoke, O. A. (2014). Testosterone promotes glucose intolerance, lipid disorder and oxidative stress in type 1 diabetic rats. *Journal of Basic Clinical Physiolology and Pharmacology,* 25:13-20

Olfert, E. D., Cross, B. M. and McWilliams, A. A. (1993) Guide to the care and use of experimental animals. *2nd ed. Ottawa, Canada: Canadian Council on Animal Care;* p. 82-93.

Orororo, O. C., Asagba, S. O., Oghri, E. and Egbune Egoamaka Oliseneku (2017) Comparative effect of garden egg, carrot and oat on biochemical parameters in cadmium exposed rats. *African Journal of Biochemistry Research*, 12(3): 28-34.

Orororo, O. C., Efekemo, O. and Udi, O. A. (2022). Changes in liver histology, hematological parameters and lipid profile of cadmium-exposed rats treated with combined leaf extract of *Vernonia amygdalina* and *Occimum gratissimum. Asian Journal of Medicine and Health;* 20(11):195-203

Orororo, O. C., Mordi, J. C., Opute, U. A., Efejene, I. O., Egbune, E. O., Busari, A. A., Badmos, K., Obadiah, C. C. and Akinshipo, W. A. (2023). Black seed oil-induced amelioration of renal dysfunction in a rat model of diabetes mellitus and periodontitis. *Tropical Journal of Natural Product Research*, 7(7):3524-3531.

Osawa T and Kato Y. (2005). Protective role of antioxidative food factors in oxidative stress caused by hyperglycemia. *Annals of New York Academy of Science,* 1043:440–451.

Ozdemir, H., Kara, M. I., Erciyas, K., Ozer, H. and Ay, S. (2012). Preventive effects of thymoquinone in a rat periodontitis model: a morphometric and histopathological study. *Journal of Periodontal Research,* 47: 74-80.

Page, R. C. and Kornman, K. (2000). The pathogenesis of human periodontitis: An introduction. *Periodontology,* 14:9-11.

Papapanou, P.N., Sanz, M., Buduneli, N., Dietrich, T., Feres, M., Fine, D.H., Flemmig, T.F., Garcia, R., Giannobile, W.V. and Graziani, F. (2018). Periodontitis: Consensus report of workgroup 2 of the 2017 world workshop on the classification of periodontal and peri-implant diseases and conditions. *Journal of Periodontology*, 89 (Suppl. S1): S173–S182.

Preshaw P.M., Alba L.A., Herrera D., Jepsen S., Konstantinidis A., Makrilakis K. and Taylor R. (2012). Periodontitis and diabetes: a two-way relationship. *Diabetologia*, 55:21–31.

Rajasekar, A. and Mathew, M. G. (2021). Prevalence of periodontal disease among individuals between 18-30 years of age: A retrospective study. *Annals of Medical Health Science Research*, 5:25-35.

Reitman, S. and Frankel, S. A. (1957). Colomentric method for determination of serum glutamic oxalacetic acid and glutamic pyruvic transaminases. *American Journal of Clinical Pathology*, 28: 56–63.

Salvi, G. E., Carollo-Bittel, B. and Lang, N. P. (2008). Effects of diabetes mellitus on periodontal and peri-implant conditions:Udate on associations and risks. *Journal of Clinical Periodontology,* 35:398–409.

Shaterzadeh-Yazdi, H., Noorbakhsh, M.F., Hayati, F., Samarghandian, S. and Farkhondeh, T. (2018). Immunomodulatory and anti-inflammatory effects of thymoquinone. *Cardiovascular and Hematological Disorder Drug Targets*, 18:52–60.

Shori, A.B, (2015). Camel milk as a potential therapy for controlling diabetes and its complications: A review of in vivo studies. *Journal of Food and Drug Analysis,* 23:609-618.

Silva, N., Abusleme, L., Bravo, D., Dutzan, N., Garcia-Sesnich, J., Vernal, R., Herna´ndez, M. and Gamonal, J. (2015). Host response mechanisms in periodontal diseases. *Journal of Applied Oral Science,* 23, 329–355.

Usman, M., Ali, M. Z., Qureshi, A. S., Ateeq, M. K. and Nisa, F. U. (2017). Short term effect of dose dependent camel milk in alloxan induced diabetes in female albino rats. *Journal of Animal and Plant Science,* 23 (3):25-38

Van Leeuwen, M. P., Slot, D. E., and Van der Weijden, G. A. (2011). Essential moils compared to chlorhexidine with respect to plaque and parameters of gingival inflammation: a systematic review. *Journal of Periodontology,* 82: 174-194.

Wahid, A., Chaudhry, S., Ehsan, A., Butt, S. and Khan, A. A. (2013). Bidirectional relationship between chronic kidney disease and periodontal disease. *Pakistan Journal of Medical Science,* 29: 211-215.

Yimer, E.M., Tuem, K.B., Karim, A., Ur-Rehman, N., Anwar, F. (2019). Nigella sativa L. (Black Cumin): A promising natural remedy for wide range of illnesses. *Evidences Based Complementary and Alternative Medicine,* 16:23-40.