

# Protective Effects of 1-Deoxynojirimycin on the Liver Histopathological Injury of Diabetic Induced Mice

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**Abstract:** Diabetes mellitus is a metabolic disorder and affects nearly 10% of the population every year. The aim of present study was to investigate the histological changes in diabetic induced Swiss albino mice, *Mus norvegicus albinus* on exposure to 1-Deoxynojirimycin. 1-Deoxynojirimycin is known to be a potent antidiabetic compound. The study was conducted on mice Swiss albino male mice, the animals were divided into six equal groups and each group comprising 5 animals. The 1st group was given distilled water and used as a control group. The 2nd and 3rd groups were normal mice they were orally administered with only 1-Deoxynojirimycin 40 mg/kg b.w. and 60 mg/kg b.w. respectively. The 4th group was used as a diabetic control which induced with Streptozotocin (STZ) to produce diabetes. The 5th and 6th groups were diabetic induced and administered 1-Deoxynojirimycin at the dose 40 mg/kg b.w. and 60 mg/kg b.w. respectively. The histological sectioning of tissue was followed as per the standard procedure. Photomicrograph of the section preparation was taken using Olympus (PM-6 Model) photomicrograph equipment. The results indicated that STZ treated mice exhibited severe liver degeneration and these changes were reversed on exposure to DNJ for a period of 30 days. The study exhibited that DNJ alleviated the hepatic histopathological changes induced by STZ.

**Keywords:** Mice, 1-Deoxynojirimycin, Streptozotocin, Necrosis, Dilated Vessels

## 1. Introduction

Mulberry not only provides food for silkworm, it also provides very useful pharmaceutical byproducts to human society. Therefore development of bioactive compounds and pharmacological studies is directly associated with improvement in the health of people. 1-Deoxynojirimycin is one of the iminosugar which suppresses the high blood glucose level that prevent the diabetes mellitus which is one of the important bioactive compound found in mulberry leaves. 1-Deoxynojirimycin is natural product which also known as azasugar, firstly reported in 1976 in the root bark of morus species [1]. DNJ and many of its derivatives such as Fagomine (1, 2-dideoxynojirimycin), Isofagomine, 4-dideoxy-1, 4-imino-D-arabinitol, 1, 4-dideoxy-1, 4-imino-D-ribitol and act as glucosidase inhibitors [2]. 1-DNJ and many of its derivatives were isolated from plants and microbes however mulberry plants contains highest percentage of 1-DNJ which is known as for its potent  $\alpha$ -glucosidase inhibition [3-10]. 1-deoxynojirimycin or aza sugar is similar to glucose in chemical structure which is a well known  $\alpha$ -glucosidase inhibitor. In the treatment of HIV, Gaucher's disease, and diabetes, 1-Deoxynojirimycin (DNJ) play a vital role which is a potent  $\alpha$ -glucosidase inhibitor [11]. STZ significantly induced biochemical alterations in liver functions [12]. Hence we interested to study the efficacy of 1-Deoxynojirimycin on diabetic and non diabetic mice.

## 2. Materials and Methods

Swiss albino mice *Mus norvegicus albinus* with body weight ranging from 25±1 grams were procured from Tamilnadu Veterinary and Animal Sciences University (TANUVAS), Chennai, India and kept in the animal house of Pondicherry University, Puducherry in different cages in an air conditioned room. The study was conducted on 30 Swiss albino male mice, the animals were divided into six equal

groups and each group comprises five animals. The 1st group was given distilled water and used as a control group. The 2nd and 3rd groups were normal mice they were orally administered with 1-Deoxynojirimycin 40 mg/kg b.w. and 60 mg/kg b.w. respectively. The 4th group used as a diabetic control which was induced with Streptozotocin (STZ) to produce diabetes. The 5th and 6th groups were diabetic induced and administered 1-Deoxynojirimycin at the dose 40 mg/kg b.w. and 60 mg/kg b.w. respectively. Diabetes was induced by giving interperitoneal injection of STZ at the doses of 55mg/kg b.w. in the 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> groups. Further the DNJ doses were given daily at 9 am for thirty days to study the efficacy of DNJ on STZ induced diabetic mice. The doses were given daily at 9 AM for thirty days. After scheduled treatment the mice were sacrificed by anesthesia. The histological sectioning of tissue was followed as per the procedure described by Humason [13]. The kidneys of mice were isolated from control and treated mice and were gently rinsed and washed with physiological saline (0.9% NaCl) to remove mucus and other debris adhering to them. They were fixed in Bauin's fluid (75 ml saturated aqueous picric acid, 25 ml 40% formaldehyde and 5 ml glacial acetic acid) for 24 hrs. The fixative was removed by washing through running tap water for overnight. Then the tissues were processed for dehydration. Ethyl alcohol was used as the dehydrating agent. Then the tissues were cleared in methyl benzoate and embedded in paraffin wax. Sections of 5 $\mu$  thickness were cut using 'SIPCON' rotatory microtome. The sections were stained with Harris hematoxylin (Harris, 1900) and counter stained with eosin, dissolved in 95% alcohol. After dehydration and cleaning, the sections were mounted in canadabalsam. Photomicrographs of the section preparation were taken using Olympus (PM-6 model) photomicrograph equipment.

### 3. Results and Discussion

Microscopic study of liver tissues revealed interesting pathological changes in the STZ and DNJ treated mice. In the 1st (Control) group the liver of mice comprises a continuous mass of hepatic cell (hepatic parenchyma cells) arranged in cords. The hepatocytes are large in size and nucleus is centrally placed. Central vein and sinusoids are found in hepatic mass (Fig. 1). Changes were observed in the liver of 2nd, 3rd, 4th, 5th and 6th groups of mice when compared to control. In the 2nd group (normal mice with DNJ 40mg/kg b.w) and 3rd group (normal mice with DNJ 60mg/kg b.w.) groups (Fig. 2 & Fig. 3), appreciable changes were not noticed. In the 4th group (Diabetic Control) (Fig. 4) central veins were dilated and the liver cells exhibited focal necrosis, blood sinusoids also dilated coagulated blood vessels. Since liver accumulates most of the toxic substance for detoxification, greater accumulation of STZ in 4th group might have resulted in extensive degeneration of the structure of liver, may be due to the failure of detoxification mechanisms. The most significant changes observed in liver of mammals after STZ administration are cell necrosis, dilation of central veins, sinusoids rupture in the vein, hyperplasia and vacuolation [14 and 15]. In the groups 5th and 6th (Fig. 5 & Fig. 6), no conspicuous changes were observed except mild necrosis. The present study has shown that STZ exert toxic effects on liver tissue. It is likely that structural alterations observed in the present study are consequent of the vascular STZ extravasations and tissue retention. Alterations observed in liver tissue in the present study are similar to the report of Mohan Rao [16] who reported that diabetic untreated rats exhibited degenerative liver with severe congestion of central vein, hemorrhages in the sinusoidal spaces, and granular appearance of the hepatocytes with cloudy swelling of nucleus. The findings have been further substantiated by biochemical disorders observed in the liver of mouse treated with STZ. The disarranged liver cord, vacuolation in hepatic cells, dilated sinusoids followed by the shrinkage of hepatocytes and dissolution of laminar structure suggest that the depilation in its glycogen reserves. Corresponding to the cellular damage to the liver tissue the decrease in proteins, increases in ammonia, impaired oxidative metabolism reflect the potent toxicity of the STZ to liver. On the other hand the diabetic control with 40 mg/kg b.w. and 60 mg/kg b.w. DNJ for a period of 30 days treatment also produced histopathological changes in the 5th and 6th groups of mice. However the net degree of damage to the liver in the group 5th is relatively less than the damage caused by STZ. Further the higher dose of DNJ 60 mg/kg b.w did not caused severe structural damage to the liver in the 6th group. This difference could be due to the high amount of DNJ accumulated in the liver. It is therefore suggested that the DNJ is beneficial in alleviating the STZ toxicity only if the DNJ treatment through oral feeding is relatively high.

### 4. Conclusions

Histopathological studies of liver on treatment with STZ and DNJ revealed histological alterations in liver. STZ induced extensive degeneration of the structure of liver. The degenerative results caused by STZ were reversed and exhibited protective effect in liver on exposure to DNJ. The

higher dose of DNJ exhibited more protective effect against STZ. Such efficiency remains to be demonstrated in field conditions.

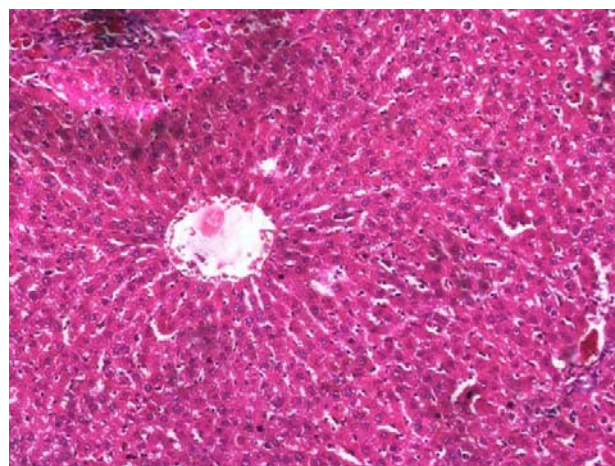


Figure 1: Transverse section of the liver of the Control Mouse (H&Ex450)

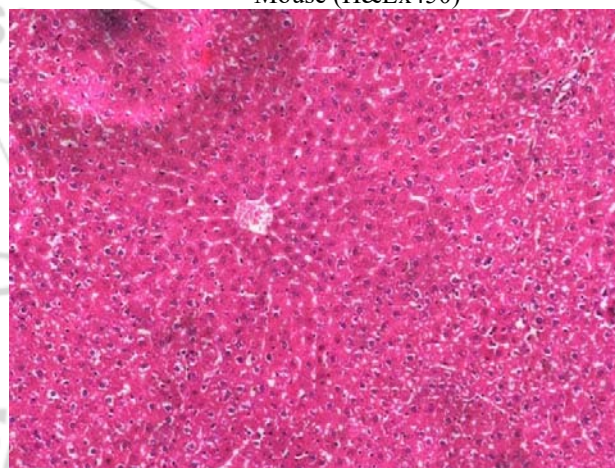


Figure 2: Transverse section of the liver of the Normal + 40 mg DNJ treated Mouse (H&Ex450)

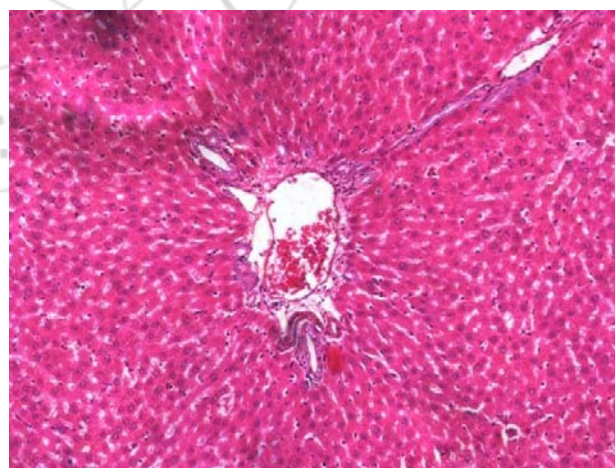
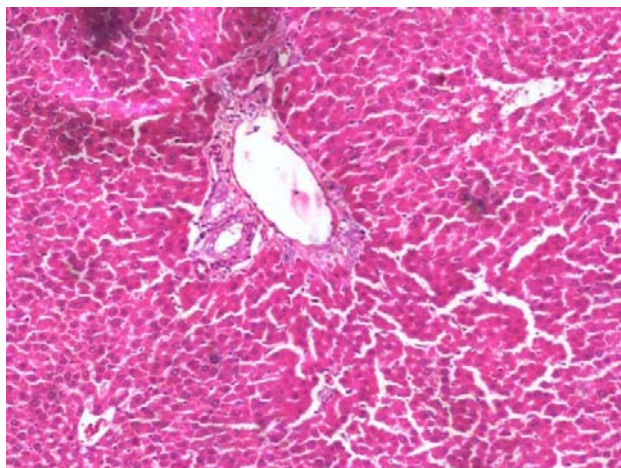
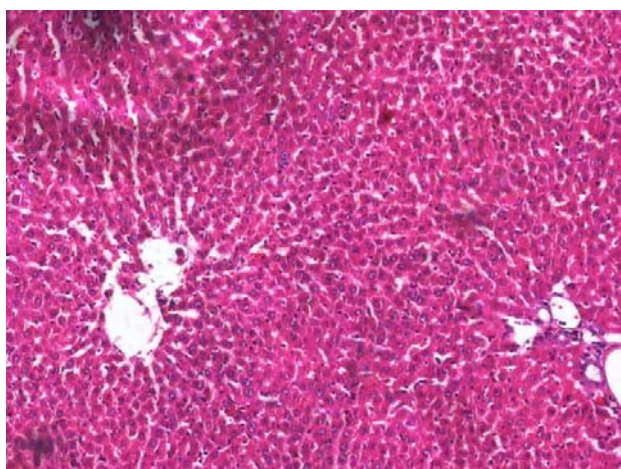


Figure 3: Transverse section of the liver of the Normal + 60 mg DNJ treated Mouse (H&Ex450)

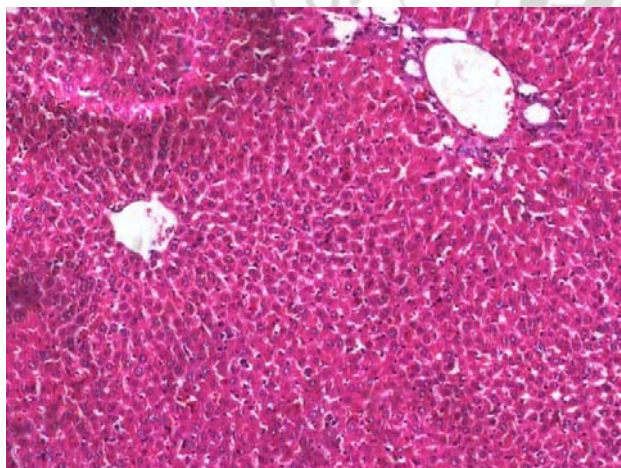




**Figure 4:** Transverse section of the Diabetic control Mouse on exposure to STZ (H&Ex450)



**Figure 5:** Transverse section of the Diabetic + 40 mg DNJ treated Mouse on exposure to STZ (H&Ex450)



**Figure 6:** Transverse section of the Diabetic + 60 mg DNJ treated Mouse on exposure to STZ (H&Ex450)

## 5. Acknowledgments

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