

Protective Effect of *Zingier officinale* on Spleen of Diabetic Guinea Pig

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ABSTRACT: *Diabetes mellitus is a chronic metabolic disease, which is associated with hyperglycaemia. This hyperglycaemia is result of defective insulin secretion or function. Traditional plant medicines or herbal formulations might offer a natural key to unlock complications of diabetes. Antioxidants play an important role to protect against damage by reactive oxygen species and their role in diabetes has been evaluated. Ginger has been used to treat a number of medical conditions, including those affecting the digestive tract such as dyspepsia, flatulence, nausea and abdominal pains etc. The present study is designed to evaluate effect of *Zingier officinale* on SGPT, SGOT, urea, uric acid and histological parameters of spleen of diabetic guinea pig. The 'treatment' groups received alloxan 150 mg/kg b.w by intra-peritoneal method once followed by eight weeks administration of aqueous extract of rhizome of *Zingier officinale* 200 mg/kg/b.w/day. SGPT, SGOT, Urea and Uric acid level were increased many folds in diabetic group of guinea pig. Glucose level were also increased many folds. Red pulp and white pulp of spleen show vacuolisation and clustered nuclei in diabetic guinea pig. While ginger administration causes marked restoration in glucose level. SGPT, SGOT, Urea and Uric acid were restored to greater extent. Marginal zone and germinal centre was degenerated. Red pulp was also restored effectively in ginger administered group. These finding indicates that *Zingier officinale* plays protective role on Liver Function Test, Kidney Function Test and spleen of diabetic guinea pig.*

Keywords: *Diabetes, Alloxan, Ginger, Marginal zone, Red pulp*

I. INTRODUCTION

Diabetes mellitus is a chronic metabolic disease, which is associated with hyperglycaemia. This hyperglycaemia is result of defective insulin secretion or function. The prolonged hyperglycaemia accompanying diabetes causes tissue damage, which results in degenerative complications in many organs including the kidney, heart, muscles, eye and many other organs (Adequate, 2001). The pancreas is well innervated by autonomic nerves rich in different types of neuropeptides including vasoactive intestinal polypeptide and NPY;2 galanin, CGRP, cholecystokinin (Adequate,1999) and leucine-enkephaline (Adeghate et al, 1996). Many neuropeptides such as galanin co-localize with hormones of pancreatic beta cells (Adeghate and Ponery, 2001). A role for glucose or glucose signalling has never been demonstrated, and in some cases, the onset of beta cell proliferation occurs prior to the onset of frank hyperglycaemia (Edvell and Lindstrom, 1999). Evidence for glucose-driven beta cell proliferation in diabetic humans is sparse (Butler et al, 2007).

The plants are used in India has about 45,000 plant species and several thousands have been claimed to possess medicinal properties (Grover et al, 2002). The active principles of many plant species are isolated for direct use as drugs, lead compounds or pharmacological agents (Fabricant and N. R, 2001). Traditional plant medicines or herbal formulations might offer a natural key to unlock diabetic complications (Nammi et al., 2003). Antioxidants play an important role to protect against damage by reactive oxygen species and their role in diabetes has been evaluated. Many plant extracts and products were shown to possess significant antioxidant activity (Sabu and Kuttan, 2002). Ginger is widely used herbal medicine for the treatment of diseases, including those affecting the digestive tract (Borelli et al., 2004). Ginger has been used to treat a number of medical conditions, including those affecting the digestive tract (Capasso et al, 2000, Capasso et al, 2003) such as dyspepsia, flatulence, nausea and abdominal pains etc.

The present study is designed to evaluate effect of *Zingier officinale* on SGPT, SGOT, urea, uric acid and histological parameters of spleen of diabetic guinea pig.

II. MATERIALS AND METHODS

Animals: The guinea pig (*Cavia porcellus*) was reared in our laboratory. The age group of guinea pig selected for the study was 12 weeks old with 450 ± 30 gm. b.w.

Chemicals: Alloxan, manufactured by Excel India Pvt. Ltd., Mumbai with EC 35% was utilized for the experiment.

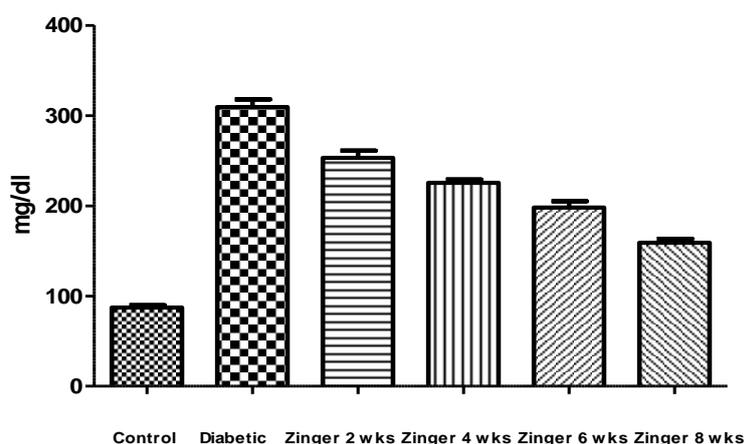
Aqueous rhizome extract of *Zingier officinale* is administered orally to diabetic group of guinea pig. Fresh rhizome of *Zingier officinale* was purchased from local herbal store in Patna, India. The identity of the rhizome of *Zingier officinale* was confirmed by Dr. Ramakant Pandey (Botanist), Department of Biochemistry, Patna University, Patna, Bihar, India.

Study groups & sampling: The control group of 6 guinea pig received distilled water as drinking water. The 'treatment' groups (n=6) received alloxan 150 mg/kg b.w by intra-peritoneal method once followed by eight weeks administration of aqueous extract of rhizome of *Zingier officinale* (200 mg/kg/b.w/day). Animals were sacrificed after the scheduled treatment. Serum was collected for SGPT, SGOT, Urea, Uric acid and glucose estimation. The spleen from all the animals were removed and washed three times in isotonic saline (0.85 v/w %) and fixed in neutral formalin for Light Microscope (LM) study.

III. RESULTS

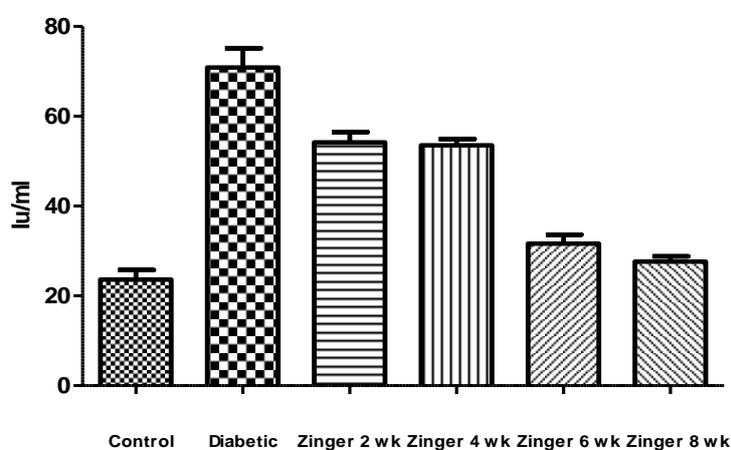
Graph: I

Glucose Level in Different Group of Guinea pig



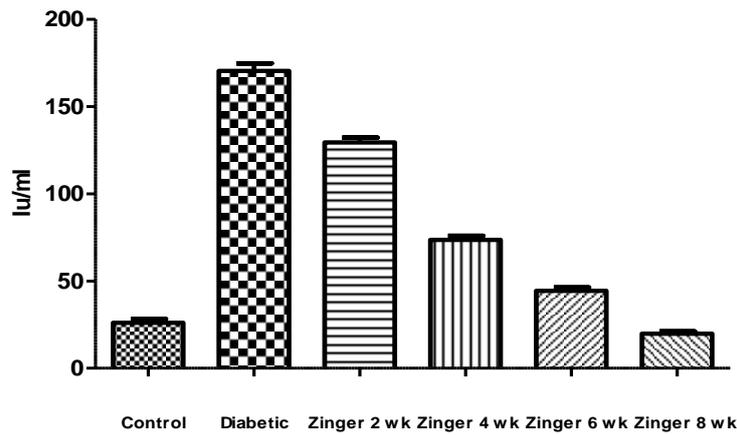
Graph: II

SGPT Level in different Group of Guinea pig



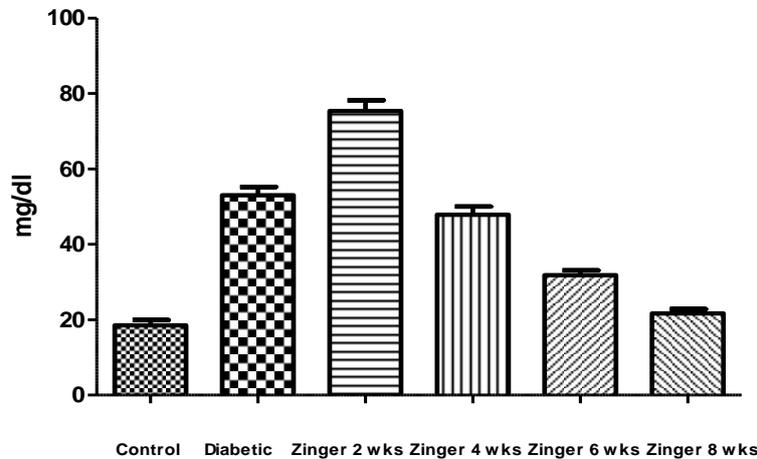
Graph: III

SGOT Level in different Group of Guinea pig



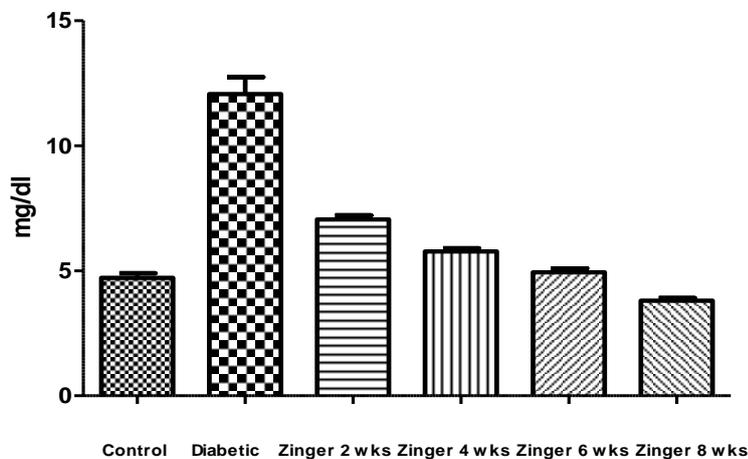
Graph: IV

Urea Level in Different Group of Guinea pig



Graph: V

Uric acid Level in Different Group of Guinea pig



Glucose level in control guinea pig was 91 mg/dl. In diabetic group of guinea pig it was 302 mg/dl, while in *Zingier* 2 weeks, 4 weeks, 6 weeks and 8 weeks administered group it was 264 mg/dl, 241 mg/dl, 216 mg/dl and 165 mg/dl gradually (Graph: I). SGPT level in control guinea pig was 24 IU/ml. In diabetic group of

guinea pig it was 72 IU/ml, while in *Zingier* 2 weeks, 4 weeks, 6 weeks and 8 weeks administered group it was 56 IU/ml, 54 IU/ml, 33 IU/ml and 28 IU/ml gradually (Graph: II). SGOT level in control guinea pig was 22 IU/ml. In diabetic group of guinea pig it was 171 IU/ml, while in *Zingier* 2 weeks, 4 weeks, 6 weeks and 8 weeks administered group it was 137 IU/ml, 79 IU/ml, 48 IU/ml and 18 IU/ml gradually (Graph: III). Urea level in control guinea pig was 18 mg/dl. In diabetic group of guinea pig it was 56 mg/dl, while in *Zingier* 2 weeks, 4 weeks, 6 weeks and 8 weeks administered group it was 77 mg/dl, 51 mg/dl, 34 mg/dl and 26 mg/dl gradually (Graph: IV). Uric acid level in control guinea pig was 4.78 mg/dl. In diabetic group of guinea pig it was 13 mg/dl, while in *Zingier* 2 weeks, 4 weeks, 6 weeks and 8 weeks administered group it was 7.68 mg/dl, 6.05 mg/dl, 5.10 mg/dl and 4.73 mg/dl gradually (Graph: V).

Spleen of control group of guinea pig shows normal red pulp and white pulp. Marginal zone is distinct. Germinal centre is well differentiated (Figure: 1). Spleen of diabetic guinea pig shows vacuolated red pulp and white pulp. Clustered nuclei were observed in marginal zone. Degenerated cytoplasm was observed in germinal centre. Central artery was also degenerated (Figure: 2). Spleen of diabetic guinea pig followed by two weeks administration of *Zingier officinale* show vacuolization in marginal zone. Clustered nuclei were observed in red pulp. Degenerated nuclei were observed in venous sinus. Vacuolated spaces were also observed in white pulp (Figure: 3). Spleen of diabetic guinea pig followed by four weeks administration of *Zingier officinale* show restoration in marginal zone. White pulp show restoration in nuclear material. Less restoration were observed in red pulp (Figure: 4). Spleen of diabetic guinea pig followed by eight weeks administration of *Zingier officinale* show more restoration in marginal zone. White pulp was also restored in their structure. Restoration in nuclei and cytoplasmic material of red pulp were observed with few vacuolated spaces. Trabecular veins were distinct (Figure: 5).

IV. DISCUSSION

Diabetes mellitus is the major endocrine disorder (Burke et al, 2003), responsible for renal failure, blindness or diabetic cataract (Thylefors, 1990), poor metabolic control (Donnelly et al., 2000) increased risk of cardiovascular disease including atherosclerosis and AGE products (Yokosuka and Nakagawa, 2004). Oxidative stress is a condition associated with increased rate of cellular damage induced by oxygen and oxygen-derived oxidants commonly known as reactive oxygen species. In a normal situation, plasma contains antioxidant mechanisms, which are likely to quench these ROS and protect against any likely damage (Gil-Del Valle et al., 2005). In present study SGPT, SGOT, Urea and uric acid were increased in diabetic group of guinea pig. While glucose also increased many folds.

Oxidative stress in DM was thought to be a result of free radicals generated during autoxidation of glucose (Miyata et al, 1999). Increased levels of ROS in type 2 DM was implicated to contribute to a hypercoagulable state (Collier et al, 1992) and most recently, evidence was provided for the accumulation of oxidation products prior to the development of diabetes (Matteucci and Giampietro, 2000). The causes of enhanced free radical production are hyperglycemia and hyper insulinemia (Hammes et al, 1997). Red pulp and white pulp show frequent vacuolization with degenerated nuclei.

Ginger is one of the most commonly consumed herbs with an array of applications in traditional medicines like Chinese medicine, Ayurveda, and Unani-Tibb (Rong et al., 2009). In ginger administered group of guinea pig SGPT, SGOT, uric acid and urea is restored effectively. It is generally considered safe and it possesses various pharmacological activities including cardiovascular protection, antioxidant, anti-inflammatory, glucose lowering, anti-cancer activities, etc. (Shukla and Singh, 2007; Nicoll and Henein, 2009). While glucose is also restored to greater extent.

V. CONCLUSION

Thus it is concluded from entire study that SGPT, SGOT, Urea and Uric acid level were increased many folds in diabetic group of guinea pig. Glucose level were also increased many folds. Red pulp and white pulp of spleen show degenerative changes in diabetic guinea pig. While ginger administration causes marked restoration in glucose level. SGPT, SGOT, Urea and Uric acid were restored to greater extent. Marginal zone and germinal centre is also degenerated. Red pulp was restored effectively in ginger administered group. These finding indicates that *Zingier officinale* plays protective role on Liver Function Test, Kidney Function Test and spleen of diabetic guinea pig.

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REFERENCES

- [1]. Adequate E., (2001): Diabetes mellitus-multifactor in aetiology and global in prevalence. Archives of Physiology and Biochemistry; 109: 197-199.

- [2]. Adequate E., (1999): Distribution calcitonin-gene-related peptide, neuropeptide-Y, vasoactive intestinal polypeptide, cholecystokinin-8, substance P and islet peptides in the pancreas of normal and diabetic rat. *Neuropeptides*; 33: 227-235.
- [3]. Adegate E., Ember Z., Donath T., Pallot D.J. and Singh J., (1996); Immuno-histochemical identification and effects of pancreastatin, leucine-enkephaline and galanin in the porcine pancreas. *Peptides*; 17: 503-509.
- [4]. Adegate E. and Ponery A.S. (2001). Large reduction in the number of galanin-immunoreactive cells in pancreatic islets of diabetic rats. *Journal of Neuroendocrinology*; 13: 706-710.
- [5]. Edvell A. and Lindstrom P. (1999). Initiation of increased pancreatic islet growth in young normoglycemic mice. *Endocrinology* 140(2): 778-83.
- [6]. Butler P.C., Meier J.J., Butler A.E., Bhushan A. (2007); the replication of beta cells in normal physiology, in disease and for therapy. *Nat ClinPractEndocrinolMetab*; 3(11): 758-68.
- [7]. Grover J. K. et al, (2002) *J Ethnopharmacol.*, 81:81 [MID: 12020931].
- [8]. Fabricant D. S. & N. R (2001). *Farnsworth, Environ Health Perspect*, 109:69 [PMID: 11250806]
- [9]. Nammi S. et al., (2003), *BMC Complement Altern Med.*, 3:4 [PMID: 12950994]
- [10]. Sabu M. C. & Kuttan. R.(2002). *Ethnopharmacol.*81:155 [PMID: 12065146].
- [11]. Borelli F., Capasso R., Pinto A., Izzo A.A. (2004). Inhibitory effect of ginger on rat ileal motility in vitro. *Life Sc* 2004; 74: 2889-96.
- [12]. Capasso F (2000). *Toxicology and clinical pharmacology of herbal products*. Totowa, New Jersey (USA): Humana Pres.; 123-129.
- [13]. Capasso F., Gaginella T.S., Grandolini G., Izzo A.A. (2003). *Phytotherapy.A quick reference to herbal medicine* SpringerVerlag, Heidelberg.
- [14]. Burke, J. P. et al,(2003), *Diabetes Care*, 26:7 [PMID: 12832302].
- [15]. Thylefors B., (1990), *The WHO program for the prevention of blindness*. *Int. J. of Ophthal.*14:211.
- [16]. Donnelly R., et. al.(2000), *BMJ*, 320:1062 [PMID: 10764371].
- [17]. Gil-Del Valle, L., Milian Lde, L., Toledo, A., Vilaro, N., Tapanes, R. and Otero, M.A. (2005). Altered redox status in patients with Diabetes Mellitus Type I. *Pharmacol. Res.* 51(4), 375-380
- [18]. Miyata T, van Ypersele de Strihou C, Kurokawa K, Baynes JW (1999) : Origin and significance of carbonyl stress in longterm uremic complications. *Kidney International*, 55:389-399.
- [19]. Collier A, Rumley A, Rumley AG, Paterson JR, Leach JP, Lowe GDO, Small M (1992) : Free radical activity and hemostatic factors in NIDDM patients with and without micro albuminuria. *Diabetes*,41:909-913.
- [20]. Matteucci E. and Giampietro O., (2000) : Oxidative stress in families of type 1 diabetic patients. *Diabetes Care*, 23:1182-1186, 2000.
- [21]. Hammes H.B., Bartmann A, Engi L. and Wulforth P (1997): Antioxidant treatment of experimental diabetic retinopathy in rats with nicanartine. *Diabetologia*, 40:629-634, 1997.
- [22]. Rong, X., Peng, G., Suzuki, T., Yang, Q., Yamahara, J., and Li, Y. (2009). A35-day gavage safety assessment of ginger in rats. *Regul. Toxicol. Pharmacol.*54:118-123.
- [23]. Shukla, Y. and Singh, M. (2007). Cancer preventive properties of ginger: a briefreview. *Food Chem. Toxicol.* 45: 683-690.
- [24]. Nicoll, R. and Henein, M. Y. (2009). Ginger: a hot remedy for cardiovascular disease? *Int. J. Cardiol.* 131: 408-409.

PHOTO PLATE

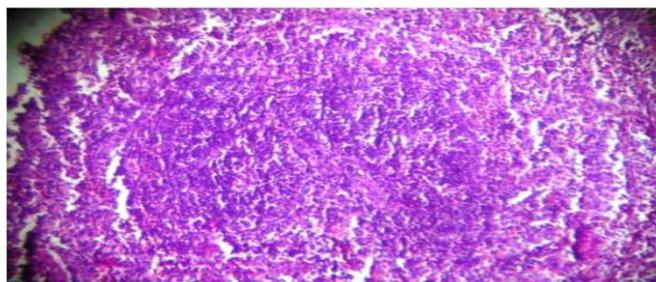


Figure-1: spleen of control guinea pig shows normal red pulp and white pulp. Marginal zone is distinct. Germinal centre is well differentiated.

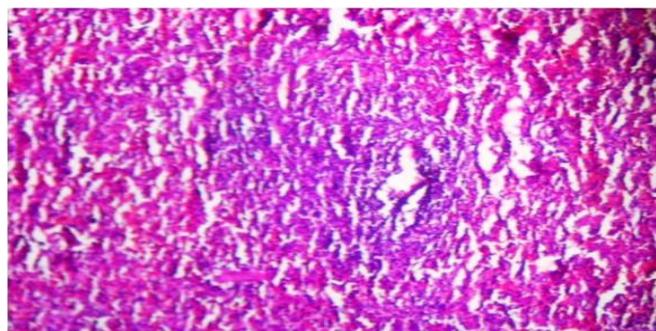


Figure-2: spleen of diabetic guinea pig shows vacuolated red pulp and white pulp. Clustered nuclei were observed in marginal zone. Degenerated cytoplasm was observed in germinal centre. Central artery was also degenerated.

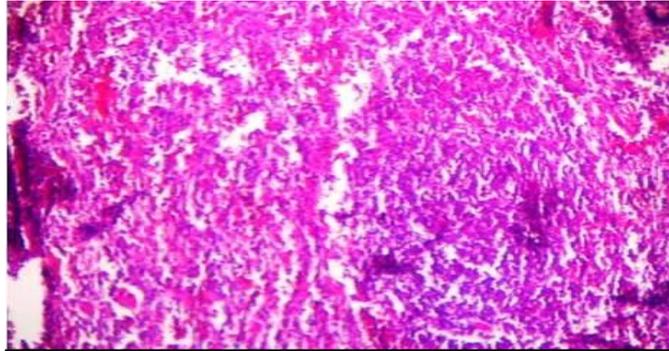


Figure-3: spleen of diabetic guinea pig followed by two weeks administration of *Zingier officinale* show vacuolization in marginal zone. Clustered nuclei were observed in red pulp. Degenerated nuclei were observed in venous sinus. Vacuolated spaces were also observed in white pulp.

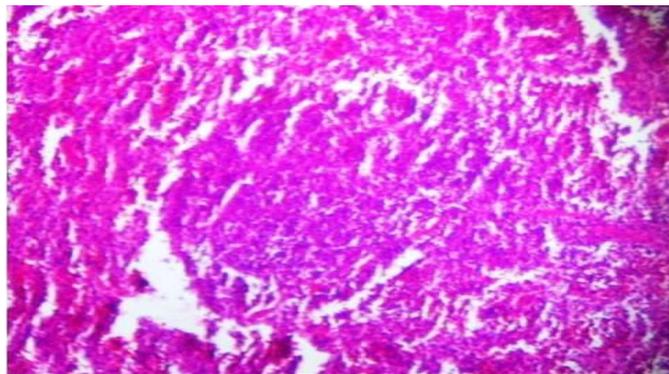


Figure-4: spleen of diabetic guinea pig followed by four weeks administration of *Zingier officinale* show restoration in marginal zone. White pulp show restoration in nuclear material. Less restoration were observed in red pulp.

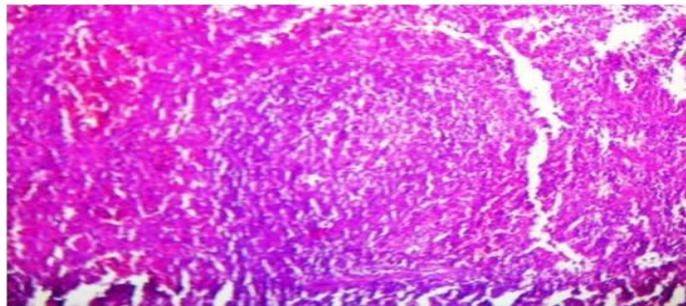


Figure-5: spleen of diabetic guinea pig followed by eight weeks administration of *Zingier officinale* show more restoration in marginal zone. White pulp was also restored in their structure. Restoration in nuclei and cytoplasmic material of red pulp were observed with few vacuolated spaces. Trabecular veins were distinct.