Journal of Pharmaceutical Sciences and Research

www.jpsr.pharmainfo.in

Effect of Nigella Sativa Seed Powder on Testosterone and LH levels in Sterptozotocine Induced Diabetes male Albino Rats.

Haseena S¹, Manjunath Aithal², Kusal K Das³, Shaik Hussain Saheb⁴.

1. Ph.D Scholar, Department of Physiology, Shri B M Patil Medical College Hospital & Research Centre, Bijapur, Karnataka, India.

2. Professor & HOD, Department of Physiology, Shri B M Patil Medical College Hospital & Research Centre, Bijapur, Karnataka, India.

3. Professor, Department of Physiology, Shri B M Patil Medical College Hospital & Research Centre, Bijapur, Karnataka, India.

4. Ph.D Scholar, Department of Anatomy, Shri B M Patil Medical College Hospital & Research Centre, Bijapur, Karnataka, India.

Abstract

Introduction -

Nigella Sativa belong to the botanical family of ranunculaceae. It has been known as black seed and its seeds are frequently used in folk medicine in Middle East and some Asian countries for the promotion of good health and treatment of many ailments. Nigella Sativa seeds has been used in traditional Iranian medicine as a natural remedy for promotes females menstruation, galactagogue, carminative, laxative and anti-parasitic properties. The present study is conducted to find out effect of Nigella Sativa seed powder on testosterone and LH levels in streptozotocine induced diabetic male albino rats.

Materials and Methods -

This work is conducted as part of Ph.D work under Department of Physiology, Shri BM patil Medical College, BLDE University, Bijapur. University ethical committee and Inistitution Animal Ethical committee are approved the work according to CPCSEA Rules. 18 rats were selected for this study and divided in to 3 groups each contains 6 rats, one group served as normal control, one group served as Diabetic control and one groups served as Treatment group with Nigella Sativa seed powder(300mg/kg BW).

Results -

Testosterone(ng/dl) level of Normal Control rats was 82.78±8.26, Diabetic rats was 41.62±7.28 and treated with Nigella Sativa rats was 71.34±6.58. LH(mIU/L) level of Normal Control rats was 0.46±0.12, Diabetic rats was 0.20±0.06 and treated with nigella sativa rats was 0.30±0.09.

Conclusion -

Compared with normal rats the level of testosterone was decreased in diabetic rats, when it is treated with Nigella Sativa Seed powder the levels of testosterone increased significantly. Compared with normal rats the level of LH was decreased significantly in diabetic rats, when it is treated with Nigella Sativa Seed powder the levels of LH not significant with other groups.

Key Words - Testosterone, LH, STZ, Diabetes, Nigella Sativa.

INTRODUCTION

The insufficient vitamins intake can cause deleterious effects on spermatogenesis and production of normal sperm[1], the sufficient consumption of vitamins and natural antioxidants can protect sperm DNA from oxidative stress and improve male fertility[2]. Vegetable oils contain several natural antioxidant constituents. Some of them such as pumpkin oil[3], celery oil[4], nigella sativa oil [5] and sunflower oil [6] were reported to possess marked antioxidant activity. Natural materials such as medical plants are wieldy accepted as feed additives. Generally, the use of chemical products may cause unfavourable side effects. Many of synthesized chemicals could cause hazards to animal or human. Different studies showed a beneficial positive effect of using Nigella Sativa seeds as feed additive in diet of ruminants. Nigella sativa is a plant of

Ranunculaceae family that grows spontaneously and widely in several Southern Mediterranean and Middle Eastern countries.

Its seed has over 100 different chemical constituents, including abundant sources of all the essential fatty acids. Although it is the oil that most often used medicinally, the seeds are a bit spicy and often used whole in cooking curries, pastries and Mediterranean cheeses[7]. Nigella sativa seeds are used extensively in traditional medicine of many countries. It has been used for treatment of many diseases owing to the reported antiviral, antiinflammatory, anti-schistosomiasis and immunomodulatory activities[8]. In recent years, the pharmacological investigations confirmed that most of the therapeutic properties of this plant are due to thymoquinone which is major active component of Nigella sativa oil[9]. Previous data suggest

that the seeds oil and thymoquinone exhibited spermioprotective effect against testes damage. Reproductive dysfunction is recognized as a consequence

of diabetes mellitus[10] represented in decrease in libido, impotence and infertility[11]. In a study poor semen quality

has also been reported in diabetic men including decreased

sperm motility, sperm concentration, increased abnormal sperm morphology[12] and reduced levels of testicular hormone[13]. Nigella Sativa seed contains a complex mixture of more than 100 compounds[14]. Most of the therapeutic properties of Nigella sativa are due to the presence of the polyphenol Thymoguinone (TQ) [15] which is the major component (28-57%) of Nigella sativa oil[16]. In addition, there are many fatty acids. The most important of which are linoleic acid (55.6%), oleic acid (23.4%) and palmitic acid (12.5%)[17]. Nigella sativa and TQ have been known by their hypoglycemic, hypolipidemic[18] and radical scavenging activity[19]. Nigella sativa oil has the ability to protect testis against oxidative stress possibly through antioxidant effects of its bioactive compounds[20]. Antioxidant property thymoquinone is attributed to the quinine structure of thymoquinone molecule[21] and the easy access to sub cellular compartments thus facilitating the ROS scavenging effect[22]. Thymoquinone was shown to inhibit nonenzymatic lipid peroxidation. This leads to decreased oxidative stress and protection of the antioxidant enzymes of testis[23]. The hypoglycemic effect of Nigella sativa oil adds more to its antioxidant effect by decreasing ROS production due to lowering glucose circulating in the blood stream. In addition, Nigella sativa oil increase the number of Leydig cells in rat testes[24] beside the presence of unsaturated fatty acids in Nigella sativa oil that stimulate 17 L-hydroxysteroid dehydrogenase activity[25] thus increasing testosterone level. The present study was conducted to study effect of Nigella Sativa seed powder on Testosterone levels in streptozotocine induced diabetic male albino rats.

MATERIALS AND METHODS

Study design - This work is conducted as part of Ph.D work under Department of Physiology, Shri BM patil Medical College, BLDE University, Bijapur. University ethical committee and Inistitution Animal Ethical committee are approved the work according to CPCSEA Rules. 18 rats were selected for this study and divided in to 3 groups each contains 6 rats, one group served as negative control, one group served as Diabetic control and one groups served as Treatment group with Nigella Sativa seed powder(300mg/kg BW), at the end of 45th day blood was collated and measured serum Testosterone and LH by kit method

Plant material – Nigella sativa seeds were grinded in to fine powder[9] with piston and mortal with help of Bapuji

pharmacy college, Davangere. Nigella sativa powder administrated orally according to study of M. Murugesan[26].

Streptozotocine – Induced diabetes -The rats were given Streptozotocine intraperitoneal injection 50mg/BW, Streptozotocine dissolved in ice-cold citrate buffer(PH 4.5)[27]. The diabetes was confirmed by measuring glucose by Code free Glucometer, the glucose level above 250mg/dl considered as diabetes, glucose levels were checked at regular periodical periods.

RESULTS

Testosterone(ng/dl) level of Normal Control rats was 82.78±8.26, Diabetic rats was 41.62±7.28 and treated with Nigella Sativa rats was 71.34±6.58. LH(mIU/L) level of Normal Control rats was 0.46±0.12, Diabetic rats was 0.20±0.06 and treated with nigella sativa rats was 0.30±0.09(Table 1).

DISCUSSION

Infertility is a complex disorder with significant medical, psychosocial and economic aspects. About 25% of couples do not achieve pregnancy within 1 year, 15% of whom seek medical treatment for infertility and less than 5% remain unwillingly childless. Infertility affects both men and women. Male causes for infertility are found in 50% of involuntarily childless couples[28]. A wide majority of medicine plants possess pharmacological principles, which has rendered them useful as curatives for numerous ailments. According to the World Health Organization (WHO) reports, 70-80% of the world population confide in traditional medicine for primary health care[29]. Plants and derivatives of plant played a key role in world health and have long been known to possess biological activity. Thirty percent of all modern drugs are derived from plants [30]. In addition, Plants have a long folklore of use in aiding fertility, including fertility-enhancing properties and aphrodisiacal qualities[31, 32].

The testes, epididymis and other reproductive organs are structurally and physiologically dependent upon the testosterone and other androgens. Testosterone stimulates growth and secretary activity of the reproductive organs[33,34]. LH stimulates the production of testosterone in Leydig cells, which in turn may act on the Sertoli and peritubular cells of the seminiferous tubules and indirectly stimulates spermatogenesis via testosterone. In our study the testosterone was decreased in diabetes induced rats and at the same time LH was decreased, in nigella sativa treated rats the testosterone levels are increased significantly but LH levels were not significant change. Our results are in agreement with studies of Mukhalad AM et, al.[35], Gokçe A et.al.[36], Rahmatollah Parandin[37].

Table 1. One way regults of Tagtagtanana(ng/dl)and I II (mI

| Table 1. One way results of Testosterone(ng/di)and LH (miU/L) | | | | | |
|---|----------------------------------|----------------------------------|------------------------------------|--------|------|
| | Group 1 | Group 2 | Group 3 | | |
| Parameter | Normal Rats- | Diabetic Rats- | Diabetic Rats – Nigella | F | P |
| | Control | Control | sativa seed powder | | |
| Testosterone(ng/dl) | 82.78 <u>+</u> 8.26 ^a | 41.62 <u>+</u> 7.28 ^b | 71.34 <u>+</u> 6.58 ^{a,c} | 33.625 | .000 |
| LH(mIU/L) | 0.46+0.12 a | 0.20+0.06 b | 0.30+0.09 a,b | 7.088 | .000 |

The difference between groups P<0.05 considered as significant.

According to Phytochemical analytical study of Nickavar B, indicated the rich presence of unsaturated fatty acids (Linoleic acid 55.6%, Oleic acid 23.4%, Palmitic acid 12.5%, Stearic acid 3.4% and else.) in nigella sativa seeds[38]. In study of Fellner et al, the supplementation of rats diets with oils rich in polyunsaturated fatty acids, such as Linoleic acid has positively influenced reproductive functions[39]. The study of Gromadzka-Ostrowska et al shown that the unsaturated fatty acids stimulate the activity of 17 β-hydroxysteroid dehydrogenase, the most important key enzyme in the testosterone biosynthesis pathway[40]. The study of Samir Bashandy found that administration of Nigella Sativa oil to hyperlipidemic rats improved their reproductive efficiency and produced additional protection against hyperlipidemia induced reduction in fertility[41]. According to study of Al-Ali A et al. Thymoguinone is the major active component derived from Nigella sativa and reported that many of the pharmacodynamic effects Nigella sativa are due to Thymoquinone[42]. The study of Gokce et confirmed that Thymoquinone treatment has protective effects on testicular parameters. The present study results are correlating with previous studies and confirmed that the testicular favouring results due to thymoquinone content in nigella sativa seed. So the nigella sativa seed may be used for increasing testicular activity.

REFERENCE

- Mosher, W.D. and Pratt, W.F. Fecundity and infertility in the United States: incidence and trends. J. Ferti. Steril. 1991;56:192-193.
- Jedlinska M, Bomba G, Jakubowski K, Rotkiewicz T, Jana B, Penkowski A. Impact of oxidative stress and supplementation with vitamins E and C on testes morphology in rats. J. Reprod. 2006;52: 203-209.
- Al-Zuhair, H, Abd EL-Fattah, A.A. and Abd El Latif: Efficacy of simvastatin and pumpkin seed oil in the management of dietaryinduced hypercholesterolemia. Pharmacol. Res. 1997;35(5):403-408.
- Momin, R.A. and Nair, M.G. Antioxidant, cyclooxygenase and topoisomerase inhibitory compounds from Apium graveolens seeds. Phytomed. 2002;9(4): 312-318.
- Shalaby, M.A. and Olfat M. Khater. Hepatoprotective and antioxidant effects of Nigella sativa oil on carbon tetrachloride induced liver damage in rats. J. Faculty Home Economic, Minufiya University. 2007;16:32-41.
- Di-Benedetto, R.; Attorri, L.; Chiarotti, F.; Eusepi, A. and Salvati, S. Effect of micronutrient-enriched sunflower oils on plasma lipid profile and antioxidant status in high-fat-fed rats. J. Agric. Food Chem. 2010;58(12): 5328-5333.
- Hajhashemi V, Ghannadi A and Jafarabadi H. Black cumin seed essential oil, as a potent analgesic and anti-inflammatory drug. Phytother. Res. 2004;18: 195-199.
- 8. Uz E, Bayrak O, Uz E, Kaya A, Bayrak R, Uz B and Turgut FH. Nigella sativa oil for prevention of chronic cyclosporine nephrotoxicity: an experimental model. Am., J. Nephrol. 2008;28:517-522.
- Gali-Muhtasib, H., EL-Najjar, N. and Schneider-Stock, R. The medicinal potential of black seed (Nigella Sativa) and its components. Adv Phytomed. 2006;2:133-53.
- Jamaludin, M., H. Norhisham, F.N. Chew, H.M.I. Muhd and K. Daren et al. Role of O-Lipoic Acid (ALA) on oxidative stress in sperm of streptozotocin induced diabetic rats. Rev. Global Med. Healthcare Res. 2012;3: 89-103.
- Jiang, G. Practical Diabetes Beijing. People's Health Publishing House, 1996; 295pp.
- Ramalho-Santos, J., S. Amaral and P.J. Oliveira. Diabetes and the impairment of reproductive function: Possible role of mitochondria and reactive oxygen species. Current Diabetes Rev. 2008;4: 46-54.

- Salih, B., T. Sipahi and E.O. Donmez. Ancient nigella seeds from Boyalı Höyük in north-central Turkey. J. Ethnopharmacology. 2009;124: 416-420.
- Ahmad, A., A. Husain, M. Mujeeb, S.A. Khan and A.K. Najmi et al. A review on therapeutic potential of Nigella sativa: A miracle herb. Asian Pacific J. Tropical Biomed. 2013;3: 337-352.
- Ali, B.H. and G. Blunden. Pharmacological and toxicological properties of Nigella sativa. Phytotherapy Res. 2003;17: 299-305.
- Nickavar, B., F. Mojab, K. Javidnia and M.A. Amoli. Chemical composition of the fixed and volatile oils of Nigella sativa L. from Iran. Z Naturforsch C. 2003;58: 629-631.
- Pari, L., S. Srinivasan and M. Saddiq. Preventive effect of diosmin, a bioflavonoid, on glycoprotein changes in streptozotocinnicotinamide-induced type 2 diabetic rats. Int. J. Pharmaceutical Sci. Res. 2010;1: 89-95.
- Al-Logmani, A. and T.A. Zari. Long-term effects of Nigella sativa L. oil on some physiological parameters in normal and streptozotocin-induced diabetic rats. J. Diabetes Mellitus. 2011;1: 46-53.
- El-Dakhakhny, M., N.J. Madi, N. Lembert and H.P. Ammon. Nigella sativa oil, nigellone and derived thymoquinone inhibit synthesis of 5- lipoxygenase products in polymorphonuclear leukocytes from rats. J. Ethnopharmacology. 2002;81:161-164.
- Danladi, J., S.A. Ahmed, S.P. Akpulu, G.K. Owolagba and M.U. Iduh et al. Protective effect of cool extraction of black seed (Nigella sativa) oil against CCl4-induced oxidative damages in Wistar Rats testis. IOSR J. Pharmacy Biological Sci. 2013;5: 68-74.
- Gokce, A., S. Oktar, A. Koc and Z. Yonden. Protective effects of thymoquinone against methotrexate-induced testicular injury. Hum. Exper. Toxicol. 2011;30: 897-903.
- Badary, O.A., A.B. Abdel-Naim, M.H. Abdel-Wahab and F.M. Hamada. The influence of thymoquinone on doxorubicin-induced hyperlipidemic nephropathy in rats. Toxicology. 2000;143: 219-226.
- Ismail, M., G. Al-Naqeep and K.W. Chan. Nigella sativa thymoquinone-rich fraction greatly improves plasma antioxidant capacity and expression of antioxidant genes in hypercholesterolemic rats. Free Radical Biology Med. 2010;48: 664-672.
- Mukhallad, A., M. Mohamad and H. Darka. Effects of black seeds (Nigella sativa) on spermatogenesis and fertility of male albino rats. Res. J. Medicine Med. Sci. 2009;4: 386-390.
- Gromadzka-Ostrowska, J., M. Przepiorka and K. Romanowicz. Influence of dietary fatty acids composition, level of dietary fat and feeding period on some parameters of androgen metabolism in male rats. Reprod. Biol. 2002;2: 277-293.
- M. Murugesan, M. Ragunath1, T. Prabu, S. Nadanasabapathi1, M. Sakthivel, V. Manju. Protective role of black cumin (Nigella sativa) on isoproterenol induced myocardial infarction in rats. International Journal of Pharmacology and Clinical Sciences. 2012;2:45-53.
- Lin S, Yang J, Wu G, Liu M, Lv Q, Yang Q, Hu J. Inhibitory effects of taurine on STZ-induced apoptosis of pancreatic islet cells. Adv Exp Med Biol. 2013;775:287-97.
- World Health Organization. WHO Manual for the Standardized Investigation and Diagnosis of the Infertile Couple. Cambridge: Cambridge University Press; 2000.
- World Health Organization. Traditional Medicine Strategy 2002-2005. Geneva; 2002.
- Burns MM. Alternative medicine: Herbal preparation. Clin Ped Emerg Med 2000; 1: 186-190.
- McDonald A. A Botanical Perspective on the Identity of Soma (Nelumbo nucifera Gaertn.) Based on Scriptural and Iconographic Records. New York: Botanical Garden Press; 2004.
- D'Cruz SC, Vaithinathan S, Jubendradass R, Mathur PP. Effects of plants and plant products on the testis. Asian J Androl 2010; 12: 468-479
- WHO: protocol CG-04. Preparation of alcoholic extract for bioassay and phytochemical studies (APJF/IP, 1001 A). Geneva, World Health Organization; 1983.
- Oberlander G, Yeung CH. Cooper TG. Induction of reversible infertility in male rats by oral Ornidazole and its effects on sperm motility and epididymal secretions. J Reprod Fertil 1994; 100: 551-550
- Mukhalad AM, Mohamad MJ, Darka H. Effects of black seeds (Nigella sativa) on spermatogenesis and fertility of male albino rats. Res J Med Med Sci 2009; 4: 386-390.

- Gokçe A, Oktar S, Koc A, Gonenci R, Yalcinkaya F, Yonden Z, et al. Protective Effect of Thymoquinone in Experimental Testicular Torsion. Eur Urol. 2010; 9: 586.
- 37. Rahmatollah Parandin, Namdar Yousofvand, Rostam Ghorbani. The enhancing effects of alcoholic extract of Nigella sativa seed on fertility potential, plasma gonadotropins and testosterone in male rats. Iran J Reprod Med. 2012;4:355-362.
- Nickavar B, Mojab F, Javidnia K, Roodgar Amoli MA. Chemical composition of the fixed and volatile oils of Nigella sativa L. from Iran. Z Nturforsch 2003; 58: 629-631.
- Fellner SJ, Sauer DS, Kramer JGK. Steady-state rates of Linoleic acid biohydrogenation by ruminal bacteria in continuous culture. J Dairy Sci 1995; 78: 1815-1823.
- 40. Gromadzka Ostrowska J, Przepiorka M, Romanowicz K. Influence of dietary fatty acids composition, level of dietary fat and feeding period in some parameters of androgen metabolism in male rats. Repord Biol 2002; 2: 277-293.
- Samir Bashandy AE. Effects of fixed oil of Nigella sativa on male fertility in normal and Hyperlipidemic rats. Int J Pharmacol 2007; 3: 27-33.
- Al-Ali A, Alkhawajah AA, Randhawa MA, Shaikh NA. Oral and intraperitoneal LD50 of Thymoquinone, an active principle of Nigella sativa, in mice and rats. J Ayub Med Coll Abbottabad 2008; 20: 25-27.