

Study Effects of Nigella Sativa Seeds Oil in Some Physiological Parameters in Experimental Heart Failure Induced by Ivabradine in Male Rats

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Abstract

Aim of this study was to investigate the effects of Nigella Sativa Seeds oil In some physiological parameters against heart failure Induced By Ivabradine in rats .A total of 60 male rats were used in this experiment, the rats were divided into three groups: control rats (n = 20); IBD (n = 20); IBD+ N. sativa oil (n = 20). The first group (G1) was a control group. The second group (G2) was given 10 mg/kg of IBD, while the third group (G3) was injected with 10 mg/kg IBD + oral dosage of 2.5 mg/kg N. sativa oil for three months., after three months, the blood was withdrawn from the heart. to measure the concentration of ANP, BNP, NE, A-II, ALDO, ACE, MDA, SOD, TNF- α and IL-6 the Results reveals that Levels of BNP, NE, MDA, IL-6 and TNF- α . were significantly increasead in G2 rats, with no significant difference was observed in ANP in rats in G2 compared to G1 rats . The level of ACE, A-II, SOD and ALDO in G2 rats had significantly decreased compared to that observed in G1 rats, oral administration of Nigella Sativa seeds oil (G3) showed no significant difference in ANP, BNP, NE, A-II, ALDO, ACE, MDA, SOD, TNF- α and IL-6 compared with G1 group.

Keywords: *Nigella Sativa, heart failure, Ivabradine, male, rats.*

Introduction

Ivabradine (IBD) It is a commonly used medication Lately used to alleviate the pain and treatment patients with chronic angina pectoris, chiefly applied to the Patients who suffer from sinus rhythm who cannot treated with beta blockers ¹. Many of studies have proved that treatment with ivabradine can enhance the function of global left ventricular and decrease accumulation of cardiac collagen in congestive heart failure in rats². Other studies has been reported that IBD can acts under the control of autonomic nervous system, thus it can reduce unsuitable sinus tachycardia³. Moreover, IBD has been found used as cardioprotective drug that used in regulating myocardial ischemic injury. IBD has been utilized for lowering risk of hospitalization and cardiovascular death rate in chronic heart failure patients^{4,5}. In spite of there are little data about the mechanisms that make IBD used as benefit drug for chronic heart failure there was many studies demonstrated that many drugs like IBD, ranolazine can decrease the sodium influx in heart muscle ⁶, decrease

thesize of myocardial muscle, increases the function of left ventricular, decreases arrhythmias and thereby improves myocardial ischemic injury^{7,8}. Nigella sativa (N. sativa) belong to Ranunculaceae Family, This plant is vastly used over the world as herbal medicines . N. sativa is considered as one of the most widely used plants for treatment or preventing of many diseases ⁹. The seeds and oils of this plant are also used in diverse of traditional systems of herbal medicine ¹⁰. The seeds and oils of N. sativa are also used as a good food components in many of systems such as food preservative and spice. Many studies revealed that, both seeds and oils have several biological effects such as anti-inflammatory, anticancer, antioxidant, antimicrobial, diuretics, antihypertensive, analgesics, anti-diarrheal and appetite stimulant, thus the N. sativa seeds can be used as effective food components¹¹. Therefore, in this study, our first focus was to investigate the effects of IBD on cardiac system and study the effects of N. sativa seeds oil against heart failure that induced by IBD .

Materials and Method

Experiment Design: A total of 60 male rats were used in this research, the rats were equally divided into three groups: control (n = 20); IBD (n = 20); IBD+N. sativa seeds oil (n = 20). The first group (G1) was given normal saline and considered as a control group. The second group (G2) was given intraperitoneally with 10 mg/kg of IBD as described by ¹. while the third group (G3) was injected inperitoneally with 10 mg/kg IBD+ oral dosage of 2.5 mg/kg N. sativa seeds oil for three months. After three months, 5 ml of blood was withdrawn from the heart. to measure the following parameters: concentration of atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), Noradrenaline (NE), angiotensin- II (A-II), aldosterone (ALDO), angiotensin converting enzyme (ACE), malondialdehyde (MDA), Superoxide dismutases (SOD), tumour necrosis factor-alpha (TNF- α) and interleukin-6 (IL-6) Plasma concentration of ANP, BNP, NE, A-II, ALDO, ACE, SOD, TNF- α and IL-6 were measured by ELISA using commercially available kits. The concentration of MDA was measured according to thiobarbituric acid (TBA) reaction by Muslih et al (2001) ¹²

Plant material and extractions: The Nigella sativa seeds utilized in this study were obtained from the local market of kerbala, Iraq. The seeds were manually checked up to eliminate bad ones. The oil from N sativa seeds was extracted by Soxhlet apparatus for 12 h and petroleum ether (60-80°C boiling range) was used as a solvent according to the method described by AOCS¹³. Later that the solvent was evaporated and finally the oil was obtained and stored -40C .

Statistical Analysis: SPSS program was used to analysis the results and we tested the correlation coefficient by means of the analysis of variance by complete randomized design (CRD). We used the least significant difference (L.S.D.) to show the significance of the results¹⁴.

Results and Discussion

Effects of Ivabradine and N. sativa seeds oil on level of ANP, BNP, NE, ACE, A-II and ALDO: Levels of BNP and NE were significantly higher among rats in G2, While no significant difference was observed in ANP in G2 compared to that observed in G1 rats (table 1) . The level of ACE, A-II, and ALDO in G2 rats had significantly reduced compared to that observed in G1 rats, the rats in G3 showed no significant difference in

all above parameters (table 1).

Table (1): Effect of Ivabradine and N. sativa seeds oil at the level of ANP, BNP, NE, ACE, A-II and ALDO in serum of male rats .

Groups Parameters	(G1)	(G2)	(G3)
ANP	1.68 0.48±	1.92 0.51±	1.71 0.56 ±
BNP	1.39 0.14±	* 2.46 0.22±	1.43 0.17±
NE	6.45 0.11±	* 6.73 0.23±	6.48 0.16±
ACE	1.68 0.17±	* 1.39 0.26±	1.66 0.22±
A-II	5.46 0.32 ±	* 4.56 0.38 ±	5.41 0.28±
ALDO	11.31 4.71±	* 8.93 5.13±	11.27 3.92±

Mean ± standard error, *= significant difference

Our data prove IB caused chronic heart failure (CHF) and amplifies the concentration of BNP and NE in plasma. This results suggest the Malfunction in left ventricle may stimulate elevation of plasma concentration of BNP and NE .

The plasma concentration of BNP and NE have been investigate as a marker for diagnosis, prognosis, screening, and monitoring treatment of CHF patients the secretion of both of these parameters (NE and BNP) which under the control of sympathetic nerves was elevated with the development of CHF ¹⁵. Many of studies demonstrated that BNP level in patients with HF are associated with alters of its plasma level¹⁶. Thus, in this study, we measured the plasma level of BNP and NE in rats that administrated with IBD.

In this study we also observed that the ivabradine therapy caused a reduction in sympathetic overdrive in rats showed by reduction in plasma levels of renin-angiotensin-aldosterone system (RAAS) components, proved by reduction in plasma levels of ACE, A-II, and ALDO. The continuation of sympathetic overdrive and long activation of RAAS have been accounted as main mediators of developing HF by developing necrotic, fluid retention, apoptotic cell death and vasoconstriction.

This may be proved by increased uptake of Ca²⁺ with ivabradine administration could have also participated to improve the diastolic function of ivabradine ¹⁷.

N. Sativa have many effects on cardiac system including cardiac depressant effect, diuretic effect and calcium channels blocking effects ¹⁸. As well as, N. Sativa was improving the inflammatory effects in many organs and protect many organs from any lesion, and we have been found that the daily use of NS with diet can protect the body from exposure to the sepsis ¹⁹. Thymoquinone(TQ), the active components of Nigella sativa found in its seeds, is the active quinone used in pharmacology, which play a vital role chiefly as anti-inflammatory and analgesic factor²⁰. Many studies has been reported that thymoquinone also act as a powerful antioxidant which protects many of body organs from oxidative injury in various studies in rats ²¹. and protects cell membrane from lipid peroxidation in many tissues ²². in addition, N. Sativa seed has a many of actions such as antioxidant ²³, calcium channel blocker effects²⁴, anti-eicosanoid ²⁵ and a regulate the level of intracellular calcium in mast cells ²⁶. NS have also myocardial protective effect which could be due to its protective activity of its components such as thymoquinone, thymohydro-quinone, thymol, dithymoquinone, 4-terpineol, carvacrol and tanethole ²⁷.

Effects of Ivabradine and N. sativa seeds oil on Myocardial Cytokines and Oxidative Stress: The results of Table 2 showed a significant increase (P < 0.05) in the level of serum MDA and significant decrease in SOD in the rats injected with ivabradine (G2) the level of IL-6 and TNF- α . Were significantly increased in G2 compared with control group (G1), oral administration of 2.5 mg/kg of Nigella Sativa seeds oil lead to enhance the level of SOD, with no significant difference was observed in level of MDA, IL-6 and TNF- α in G3 compared with G1 group.

Several studies have indicated that there was many effects of ivabradine on cardiac system especially inflammation^{28,29}. Lately, some of studies proved that IBD can reduce the level of inflammatory cytokines in mice²⁸. And can modify the gene expression of inflammatory cytokine while elevate the activity of endothelial nitric oxide synthase (eNOS) in rats²⁹. Other studies suggested that, treatment with ivabradine can promote the function of left ventricle, with modifying in cytokines expression mainly TNF- α and IL-6 . This cytokines are responsible for any defect in cardiac

system³⁰. Especially TNF- α and IL-6 which stimulate cardiac dysfunction. In this research, we found that ivabradine caused cardiac inflammation by increased levels of IL-6 and TNF- α .

Table (2): Effect of Ivabradine and N. sativa seeds oil at the level of MDA, SOD, TNF- α , and IL-6 in serum of male rats

Groups Parameters	(G1)	(G2)	(G3)
MDA	0.0492 0.022±	* 0.055 0.310 ±	0.0488 0.019 ±
SOD	58.46 11.05±	* 42.62 6.05±	58.97 6.35±
TNF- α	2.19 1.59 ±	* 3.85 1.77±	2.11 1.35±
IL-6	35.24 8.17±	* 39.92 3.14±	34.97 6.14±

□ Mean ± standard error, * = significant difference

Conclusion

The results suggest that injection with Ivabradine can induce heart failure and oral administration of Nigella Sativa Seeds oil has protective effects against heart failure Induced By Ivabradine.

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Conflict of Interest: None to declare.

Ethical Clearance: All experimental protocols were approved under the College of Education for Pure Science and all experiments were carried out in accordance with approved guidelines.

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