WORLD JOURNAL OF PHARMACY AND PHARMACEUTICAL SCIENCES
SJIF Impact Factor 7.421
Volume 8, Issue 2, 1257-1262 Research Article ISSN 2278 – 4357

PROTECTIVE EFFECTS OF AQUEOUS HOT EXTRACT OF *EUGENIA UNIFLORA* LEAVES IN ISOPROTERENOL-INDUCED MYOCARDIAL INFARCTION IN MALE RATS-HISTOLOGICAL EXAMINATION

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Article Received on
21 Dec. 2018,

Revised on 11 Jan. 2019,
Accepted on 31 Jan. 2019

DOI: 10.20959/wjpps20192-13171

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ABSTRACT

In this investigation, the protective effects of aqueous hot extract of *Eugenia uniflora* leaves in Isoproterenol induced myocardial infarction in male rats was studies. The histopathological architecture of heart tissue of control and induced rats were studied. Rats were pretreated with hot aqueous extract of *Eugenia uniflora* leaves orally daily for 28 days. After treatment, Isoproterenol was injected to rats at an interval of 24 hours for two days to induce myocardial injury. The results obtained emphasized the benefits of the extract and thus augmented the urge of in vivo studies to further confirm the beneficial effect of these extract.

KEYWORDS: Myocardial injury, aqueous extract, *Eugenia uniflora*, Isoproterenol, histopathology.

INTRODUCTION

Myocardial infarction (MI) is a lethal manifestation of IHD and disability worldwide. MI can be as a proxy for the study of CAD (Coronary artery disease) in a population.[1] Normally our heart to continue its function there should be sufficient supply of blood, oxygen and nutrients which is provided by circulation. Myocardial ischemia could be explained as an irreversible damage of a particular area of the heart muscle (myocardium) due to blocked supply of blood to that particular area. Without proper treatment this condition can lead to death. MI is a part of a range of disorders called ACS (Acute coronary syndrome).

One of the major causes for MI is thrombosis or blood clot that forms inside an artery or one of its branches. Root problem for the formation of clot is atheroma (plaque).[2] Plaques or the fatty patches may gradually form over several years in one or more than one place inside the coronary arteries. MI is followed by many biochemical and pathophysiological alterations like hyperlipidemia, lipid peroxidation, thrombosis and free radical damage leading to the change in myocardium. Among the various mechanisms, the accumulation of free radical have been involved in the pathophysiology of Acute Myocardial Infarction (AMI).Oxidative stress caused by free radicals and ROS (Reactive Oxygen Species) have noted in production of lipid peroxides with decreased levels of different antioxidants which plays a serious role in the myocardial damage.

Use of animal models to study cardiac diseases helps to understand the pathogenesis of the disease and also for the development of several diagnostic techniques. Out of many well known animal models, the rat model of Isoproterenol induced myocardial necrosis is used to study cardiac dysfunction. Isoproterenol [1-(3, 4-dihydroxyphenyl) - 2-isopropyl amino ethanol hydrocholoride] (ISO) is a synthetic catecholamine and a β-adrenergic agonist which causes severe oxidative stress in the heart muscle which results in necrosis.[3] ISO stimulates lipid peroxidation and also generates free radicals which cause irreversible damage to the myocardial membrane.

Several mechanisms are presented for ISO induced myocardial infarction. When MI occurs there is an imbalance between the demand and supply of oxygen inside heart muscle which causes myocardial hyperfunction due to increase in both inotropism and chronotropism and results in hypotension in coronary bed. Also, there is an elevation of calcium ions inside the cell, this in turn activates adenylate cyclase enzyme and results in the depletion of ATP levels. Eventually there is an oxidative stress because of several metabolic products (free radicals) originated from ISO.

The involvement of oxidative stress in the cause of several acute and chronic diseases suggests antioxidant therapy as a promising solution for the treatment of these diseases. Since time immemorial people depends in nature for drugs. From prehistoric period onwards plants have been used for medicinal purposes. India has known for its rich repository of medicinal plants. Plants have an important role in welfare of human beings. Nowadays there is a great interest for natural remedies for the treatment of various ailments because of its safety and

minimal side effects. In present scenario, synthetic drugs are regarded harmful to human beings.

Eugenia uniflora, is an evergreen shrub belonging to Myrtaceae family. Various study report suggests that *Eugenia uniflora* is a functional food which contains different compounds with antioxidant, anti-diabetic, anti-inflammatory effects. Previous studies have reported the therapeutic effects of various parts of *Eugenia uniflora.* Leaves of *Eugenia uniflora* have been used for treating gout, hypertension, inflammation, digestive disorders, rheumatism, cough, hepatic diseases, sore throat, haemorrhoids and amygdalitis. *Eugenia uniflora* also have antibacterial, antifungal, antiviral and antprotozoal activities. Various metabolites like flavanoids, terpens, tannins, steroids, phenols, essential oil etc, were identified in in *Eugenia uniflora*, these metabolites contributes for the biological properities.[4-10]

Considering all available scientific background supporting the pharmacological effects of in *Eugenia uniflora* leaves, present study presents the histopathological examination of heart tissue of control and isoproterenol induced rats.

MATERIALS AND METHODS

Plant Collection and Authentication: Fresh leaves of Eugenia uniflora (Linn), Family-Myrtaceae, were collected from Wayanad district, Kerala during the month of April 2014. Taxonomic authentication was done by Dr. V.S Ramachandran, Taxonomist, Department of Botany, Bharathiar University, Coimbatore, Tamil Nadu, India.

Sample Processing: The leaves were washed, shade dried at room temperature and powered in a mixer grinder.

Hot Water Decoction: 10g of the powdered sample was dissolved in 100ml of distilled water which was boiled for one and half hours and filtered. The decoction was stored at 4 °C for further usage.

Chemicals: All chemicals used for the evaluation were in analytical grade and obtained from either Sigma–Aldrich or Merck.

Induction of toxicity: ISO was dissolved in 1 ml of normal saline and injected subcutaneously at an interval of 24 hours to rats (85mg/kg b.wt) daily for two consecutive day's i.e. 28th and 29th day to induce experimental myocardial infarction. The choice of ISO

dose was based on a pilot study for ISO dose fixation and on the results of a previous study reported by Panda and Naik, 2008.

Experimental Design: After the acclimation period animals were divided into four groups containing six animals in each group. All animals were fed with pellet and water. The experimental design is as follows,

Group I: Normal control rats fed with standard rat chow pellet and pure drinking water

Group II: Rats were administered with ISO (85 mg/kg b.wt) dissolved in normal saline subcutaneously.

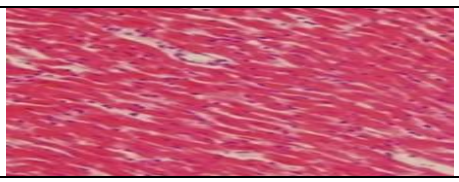
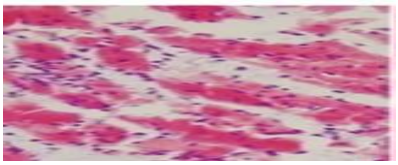
Group III: Rats were pretreated with aqueous extract of *Eugenia uniflora* leaves (200 mg/kg b.wt) and then administered with ISO (85 mg/kg b.wt) dissolved in normal saline subcutaneously.

Group IV: Administered with aqueous extract of *Eugenia uniflora* leaves (200 mg/ kg b.wt)

After the experimental regimen, heart samples were washed immediately after excision and washed in normal saline and preserved in 10% formalin. The histopathological examination of heart sample from each groups were carried out according to the standard procedure given by Dunn, 1974[12] to assess the architecture of the cell.

RESULTS AND DISCUSSION

From the year 1970s onwards many research studies have published using the isoproterenol model for MI in rats inorder to evaluate the effects of drugs**.** Studies have reported that the administration of ISO in large dose causes morphological and functional changes in the heart which leads to myocardial necrosis. The toxic effects of catecholamines due to the oxidation of hydroxyl group in catecholamines leads to conversion into quinones and formation of adrenochromes which cause cell necrosis and failure in rats heart. Highly toxic oxygen derived free radicals are produced during this stage. The histopathological examination in heart tissue of control and induced rats is shown in figure 1.

	
Group 1: Control	**Group 2: ISO induced**

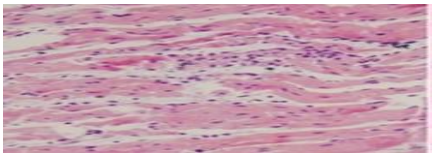
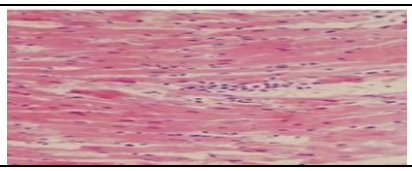
	
Group 3: ISO+extract (200 mg/kg b.wt)	**Group 4:Extract alone (200mg/kg b.wt)**

Figure. 1: The histopathological examination in heart tissue of control and induced rats.

From the figure, it is clear that group I, control animals shows myocardial fibres with normal histology. ISO treated groups (group II) shows myocardium with congested blood vessels, inflammation and extravasated RBCs. A mild lymphocytes infiltrate is observed. The animals pretreated with the aqueous extract of *Eugenia uniflora* leaves shows myocardial fibers with normal histology. The result indicates that the aqueous extract prevented the degeneration of myocardial tissue and infiltration of lymphocytes. Studies have reported that the pharmacological endogenous antioxidants has identified as very effective therapeutic approach for the diseases associated with oxidative stress. Chemically, *Eugenia uniflora* is reported to contain larhe amounts of metabolites like flavanoids, alkaloids, phenols etc which may contributye to the antioxidant activity. It is concluded that the aqueous extract has the potential to inhibit the toxic effects induced by ISO and possesses therapeutic value in the treatment of MI due to the antioxidant effect of the extract. The result obtained is in agreement with the report given by Heraldo *et al.*, 2011.[13]

REFERENCES

1. Sanchis-Gomar F, Perez-Quilis C, Leischik R, Lucia A. Epidemiology of coronary heart disease and acute coronary syndrome. Ann Transl Med., 2016; 4(13): 256.
2. Rafieian-Kopaei M, Setorki M, Doudi M, Baradaran A, Nasri H. Atherosclerosis: process, indicators, risk factors and new hopes. Int J Prev Med., 2014; 5(8): 927-4.
3. Karthick M, Prince PS. Preventive effect of rutin, a bioflavonoid on lipid peroxides and antioxidants in isoproterenol-induced myocardial infarction in rats. J Pharm Pharmacol, 2006; 58: 701-7.
4. Consolini AE, Sarubbio MG. Pharmacological effects of Eugenia uniflora (Myrtaceae) aqueous crude extract on rat's heart. J Ethnopharmacol, 2002; 81(1): 57-63.
5. Weyerstahl P, Marschall-Weyerstahl H, Christiansen C, Oguntimein BO, Adeoye AO. Volatile constituents of Eugenia uniflora leaf oil. Planta Med., 1988; 54(6): 546-9.
6. Victoria FN, Lenardão EJ, Savegnago L, Perin G, Jacob RG, Alves D, et al. Essential oil of the leaves of Eugenia uniflora L.: Antioxidant and antimicrobial properties. Food Chem Toxicol, 2012; 50: 2668-74.

7. Consolini AE, Baldini OA, Amat AG. Pharmacological basis for the empirical use of Eugenia uniflora L. (Myrtaceae) as antihypertensive. J Ethnopharmacol, 1999; 66(1): 33-9.
8. Ogunwande A, Olawore NO, Ekundayo O, Walker TM, Schmidt JM, Setzer WN. Studies on the essential oils composition, antibacterial and cytotoxicity of Eugenia uniflora L. Int J Aromatherap, 2005; 15(3): 147-52.
9. Costa P, Filho EG, Silva LM, Santhos SC, Passos CS. Influence of fruit biotypes on the chemical composition and antifungal activity of the essential oils of Eugenia uniflora leaves. J Braz Chem Soc, 2010; 21(5): 851-8.
10. Ibikunle GF, Adebajo AC, Famuyiwa FG, Aladesanmi AJ, Adewunmi CO. In-vitro evaluation of anti-trichomonal activities of Eugenia uniflora leaf. Afr J Tradit Complement Altern Med., 2011; 8(2): 170-6.
11. Santos KK, Matias EF, Tintino SR, Souza CE, Braga MF, Guedes GM, et al. Anti-Trypanosoma cruzi and cytotoxic activities of Eugenia uniflora L. Exp Parasitol, 2012; 131(1): 130-2.
12. Dunn, W.L. (1974). Hand book of histopathological and histochemical technique, 3rd edition, Red wood, Burn, Ltd.
13. Heraldo Guedis Lobo Filho., Nestor Lemos Ferreira., Rafael Bezerra de Sousa., Eduardo Rebouças de Carvalho., Patrícia Leal Dantas Lobo., José Glauco Lobo Filho. Experimental model of myocardial infarction induced by isoproterenol in rats. Rev Bras Cir Cardiovasc, 2011; 26(3): 469-76.