ejpmr, 2020,7(4), 161-168

SJIF Impact Factor 6.222

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Review Article

ISSN 2394-3211

EJPMR

IN-VIVO PHARMACOLOGICAL ACTIVITIES OF METHANOL EXTRACT OF *RHODODENDRON ARBORETUM* (LEAVES) ON EXPERIMENTAL MICE

Mohammad Moniruzzaman[1*], Mohammed Tareq Aziz[1], Md. Ariful Mondol Arif[1], S. M. Farzana[1]

[1]Department of Pharmacy, Bangladesh University, Bangladesh, Dhaka-1207.

***Corresponding Author: Mohammad Moniruzzaman**

Department of Pharmacy, Bangladesh University, Bangladesh, Dhaka-1207.

Article Received on 06/02/2020 **Article Revised on 27/02/2020** **Article Accepted on 19/03/2020**

ABSTRACT

The point of this examination was to explore the analgesic, anti-inflammatory and hypoglycemic exercises of methanol concentrate of *Rhododendron arboretum* leaves alongside the nearness of significant phytochemicals. The rough leaf concentrate of *Rhododendron arboretum* was examined for analgesic, anti-inflammatory and hypoglycemic exercises utilizing different exploratory models. Against diabetic movement was dictated by Oral Glucose Tolerance Test (OGTT). To decide the analgesic movement, acidic corrosive actuated squirming model was utilized on mice. For anti-inflammatory movement test Inflammation (paw edema) was actuated by infusing 0.1ml of 1% Carrageenan. The concentrate caused a significant portion subordinate decrease of glucose, aggravation and torments instigated by various operators utilized. The phytochemical screening demonstrated the nearness of glycoside, alkaloid, flavonoid, saponin, tannin and steroid kind of mixes. Leaf separate has calming and pain relieving impacts which might be intervened through the phytochemical constituents of the plant.

KEYWORDS: *Rhododendron arboretum*, Hypoglycemic activity, Analgesic activity, Anti-inflammatory activity, Carrageenan.

INTRODUCTION

First stage is caused with porousness increment and exudation of liquid into interstitial space, furthermore aggravation of leukocytes from blood to tissue, thirdly granuloma development. Calming tests have three stages: intense, sub intense and incessant fix process.[1] Irritation is a complex natural reaction of vascular tissue by pathogens, aggravations with redness, warm, growing and agony.[2-3] Aggravation is of two sorts: intense and ceaseless. Intense aggravation is an underlying reaction of destructive boosts and incessant is the harm to the body prostaglandins, prostacyclins and thromboxanes that are associated with irritation, torment are combined by cyclo-oxygenes (Cox).[4]

Irritation is a progression of components with contamination, injury and injury to the tissues.[5] Aggravation is started by catalyst initiation, arbiter discharge, cell relocation, liquid extravasations.[6] Aggravation discharges leukocytes against injury that blend some biomolecules and discharges during growing and redness.[7] During aggravation prostaglandin is blended and prompts irritation.[8] Continue aggravation can lead undesired wellbeing impact. Aggravation has been shown in a few illnesses including malignant growth.[9-10]

Irritation blocking specialists assume a significant job in treating pathologies related with provocative response.[11] In spite of the fact that irritation has many reasons yet instruments are same to all. The provocative specialist actuates phospholipase A2 in the site, discharge arachidonic corrosive and metabolites. Provocative middle people like cytokine, histamine, serotonin, prostaglandin increment cell porousness to the site of activity.[12] Any break right now in the decrease of middle person discharge and come back to ordinary hemodynamic condition.[13] Individuals look for clinical assistance because of agony that is unwanted physical and enthusiastic experience. For bothersome symptom of agony executioner drugs, treatment for interminable torment is general medical issue.[14] Pain relieving that lessens torment can be grouped into three classes: narcotic pain relieving (morphine), non-narcotic pain relieving (NSAIDs) and adjuvant pain relieving (which is taken for other reason however decreases torment in specific circumstances). In view of high viability, narcotic pain relieving gives maximal absense of pain.[15]

Because of symptoms (hypertension, highperglycemia, osteoporosis, cardiovascular sickness) clinical utilization of these medications experience the ill effects of hindrances.[16] Factor of finding new mixes for torment treatment has comprehended a mind boggling component of agony transmission to sensory system as a result of

having numerous receptors, protein and flagging way ways.[17] Pharmacological component is viewed as the capability of therapeutic plants for find of new mixes in the treatment of agony issue (less symptom).[18]

One of the most well-known endocrine issue is diabetes mellitus and in excess of 100 million individuals are experiencing it because of populace development, maturing, increment heftiness and physical inertia.[19-20] In India more individuals are experiencing type 2 diabetes than other nation[21-22] by ongoing measurement, around 7.8% (438 million) individuals is required to have diabetes constantly 2030.[23] A higher number of individuals are influencing by this malady because of stress, quick improvement of urban communities, way of life straightforwardness and metro life. The use of treatment of diabetes surpasses $100 billion every year.[24] In view of having reaction and significant expense, the treatment of diabetes with manufactured medications are not best but rather customary medications are best for less symptom. In any event 400 plants have against diabetic action as indicated by writing.[25] Diabetes mellitus is basic endocrine ailment have microvascular (retinopathy, neuropathy, and nephropathy) and macrovascular (coronary failure, stroke and fringe vascular illness) intricacies.[26] Because of lack of insulin individuals experience the ill effects of diabetes with high sugar level.[27] Type 2 is the most widely recognized structure wherein body doesn't appropriately utilize insulin.[28] Presently treatment of this infection with engineered tranquilize is accessible but since of having increasingly reactions the pursuit of compelling and less symptom hypoglycemic specialist is obligatory.[29] The hypoglycemic impact of a few plants utilized as against diabetic cures and the component is being contemplated. Customary meds from restorative plants offer extraordinary potential for revelation of new enemy of diabetic medications.[30]

MATERIALS AND METHOD

Drugs and Chemicals

Carrageenan was purchased from Otto chemicals, India. The standard drug Diclofenac-Na was received as a gift from Square Pharmaceuticals Limited of Bangladesh. Acetic acid, methanol and other chemicals supplied from laboratory of Bangladesh University were analytical grade.

Experimental Animals

Eight weeks-old Swice albino mice (27-30g) purchased from Jahangirnagar University, Dhaka, Bangladesh and were housed in animals cages under standard environmental conditions (22-25°C, humidity 60-70%, 12 hr light: 12 hr dark cycle). The mice were feed with standard pellet diet taken from, Jahangirnagar University, Dhaka. The animals used in this study were cared in accordance with the guidelines on animal experimentation of our institute.

Plant Materials

The flowering plant of *Rhododendron arboretum* leaves were collected from Nepal.

Drying and Grinding

The collected plants were separated from undesirable materials or plants or plant parts. They were dried in the sun for one week after cutting into small pieces. The plant parts were ground into coarse powder with the help of a suitable grinder. The powder was stored in an airtight container and kept in a cool, dark and dry place until analysis commenced.

Preparation of Plant Extract

About 430gm of powdered sample was taken in a clean, flat-bottomed glass container and soaked in 1200 ml of 90% methanol. The container with its contents was sealed and kept for a period of 10 days accompanying occasional shaking and stirring. The whole mixture then underwent a coarse filtration by apiece of clean, white cotton material. Then it was filtered through whatman filter paper. The filtrate was kept in an open space to evaporate the solvent thus crude extract was obtained. Fine powders of the flowering plant of *Rhododendron arboretum* leaves are dissolved in 90% methanol and then evaporation the solvent.

Phytochemical Screening

Phytochemical studied of methanol extract of plant material was carried out for preliminary chemical investigation for the direction of practical pharmacognosy text book.[31-33]

Anti-inflammatory Activity

Inflammation (paw edema) was induced by injecting 0.1ml of 1% Carrageenan in physiological saline into the sub plantar tissues of the left hind paw of each mouse.[34] The methanol extract of *Rhododendron arboretum* leaves rind 250mg/kg and 500mg/kg were administered orally 30 min prior to Carrageenan administration. The paw edema size was measured at 0, 1, 2, 3 & 4 hours by using dial caliper.[35] The percentage reduction of paw edema in drug treated group was compared with the control group. Diclofenac sodium (5 mg/kg p.o.) was used as reference standard. 0 hour reading was considered as an initial normal paw size. Data was collected from the paw thickness and percentage reduction of paw edema of the treated animals. Percentage reduction of paw edema was calculated by using the formula.

Anti-inflammatory activity (%) = (1-T/C) x100

Where T is the change of paw diameter in treated group and C is the change of paw diameter in control group.

Analgesic Activity

For analgesic test all mice were divided into four groups. Each group comprises of 4 mice. Control group (received 0.5% methyl cellulose, per oral), Standard Group (received Diclofenac-Na10mg/kg intraperitoneally), and *Rhododendron arboretum* leaves extract Group (received

250mg/kg and 500mg/kg extract per oral). The analgesic activity of the samples was studied using acetic acid-induced writhing model in mice. Test samples and vehicle were administered orally 30 mins before intraperitoneal administration 10ml/kg of 1% acetic acid but Diclofenac-Na was administered intraperitoneally 15 minutes before the acetic acid injection, the mice were observed for specific contraction of body referred to as "writhing" for the next 10minutes.[36] Percentage protection of acetic acid induced writhing was calculated by the formula.

Percentage protection = (Wc-Wt)/Wc x100
Where, wc is the mean values of control group and Wt is the mean values of treated group.

Anti-diabetic Activity
Oral Glucose Tolerance Test (OGTT) in diabetic mice. After fasting 16hr, diabetes was induced into mice by in intra-peritoneal injection (i. p.) of alloxan monohydrade (90 mg/kg) dissolved in saline. After 72hrs, plasma glucose levels were measured by glucometer using a blood sample from tail-vein of mice. Mice with blood sugar higher than11.5mmol/L were considered as diabetic.

All the mice were divided into 4 groups, each group containing 5 mice. The divided groups are NC (normal control), DC (diabetic control), STD (diabetic mice receiving 100mg/kg Metformin), ME (diabetic mice receiving 250mg/kg and 500mg/kg methanol extract). The mice were fasted over-night and next day blood samples were taken from all groups of animals to estimate fasting blood glucose level (0 min). All mice received 2gm/kg glucose. Without a delay extracts and was given per oral and four more blood samples were collected at 30, 60, 90 and 120 minutes intervals and blood glucose level was estimated in all the experiments by using glucometer.[37]

RESULT AND DISCUSSION

Results of Phytochemical Screening Test

Table 1: Results of Phytochemical Screening Test.

Phytochemical Group	Results
Alkaloids	+
Tannins	-
Flavonoids	+
Saponins	+
Gums	-
Steroids	+
Alkaloids	+
Glycoside	+
Acidic compounds	-
Proteins	+
Terpenoids	-

Note: + = Indicates the presence of the tested group, - = Indicates the absence of the tested group.

Results of Anti-inflammatory Activity Test

Table 2: Anti-inflammatory effects of the methanol extract of *Rhododendron arboretum* leaves extract on 1% Carrageenan induced paw edema on mice.

Group	Volume of Paw Edema (mm)					
	Blank	0 min	60 min	120 min	180 min	240 min
Control	2.98±0.10	4.40±0.11	4.28±0.06	4.18±0.07	4.10±0.07	4.08±0.06
Standard	2.75±0.10	4.83±0.02	3.88±0.06	3.50±0.04	3.05±0.05	2.83±0.11
R. arboretum 250 mg/kg	3.03±0.06	4.68±0.17	4.13±0.13	3.68±0.13	3.33±0.11	3.05±0.06
R. arboretum 500 mg/kg	2.90±0.04	4.60±0.10	3.98±0.08	3.55±0.08	3.20±0.07	2.90±0.04

Values were expressed in (Mean ± SEM) value. Each group comprised 4 animals. Control Group received normal water and standard group received 10mg/kg Diclofenac sodium. Extract group was treated with 250mg/kg and 500mg/kg of the crude extract of *Rhododendron arboreum*.

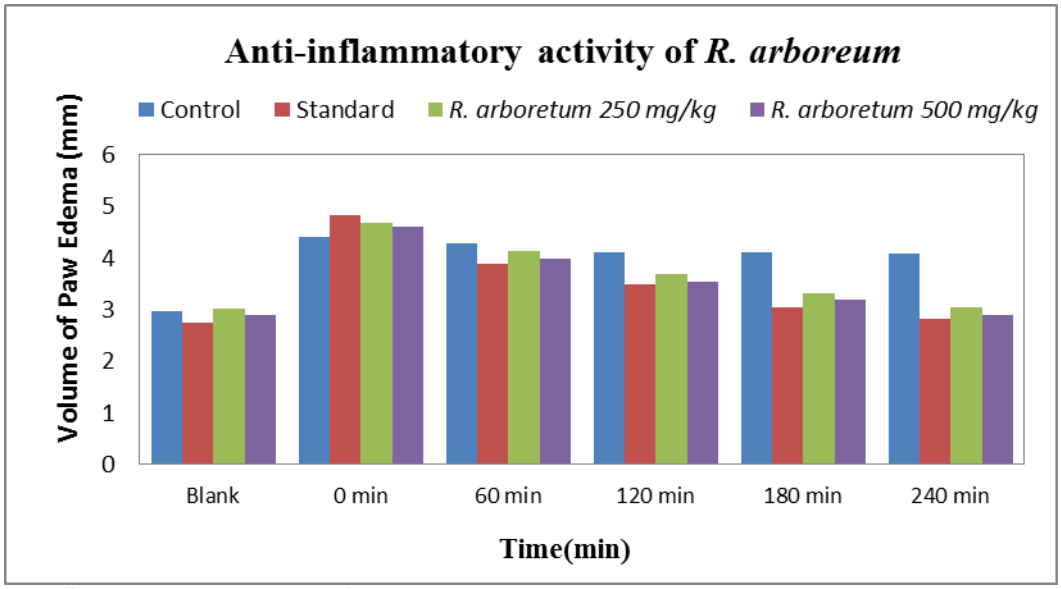

Fig 1: Effects of the methanol extract of *Rhododendron arboreum* on Carrageenan induced paw edema in mice.

Table 3: % inhibition of Paw Volume.

Group	% inhibition of Paw Volume
Control	0
Standard	84
R. arboreum 250mg/kg	80
R. arboreum 500mg/kg	81.17

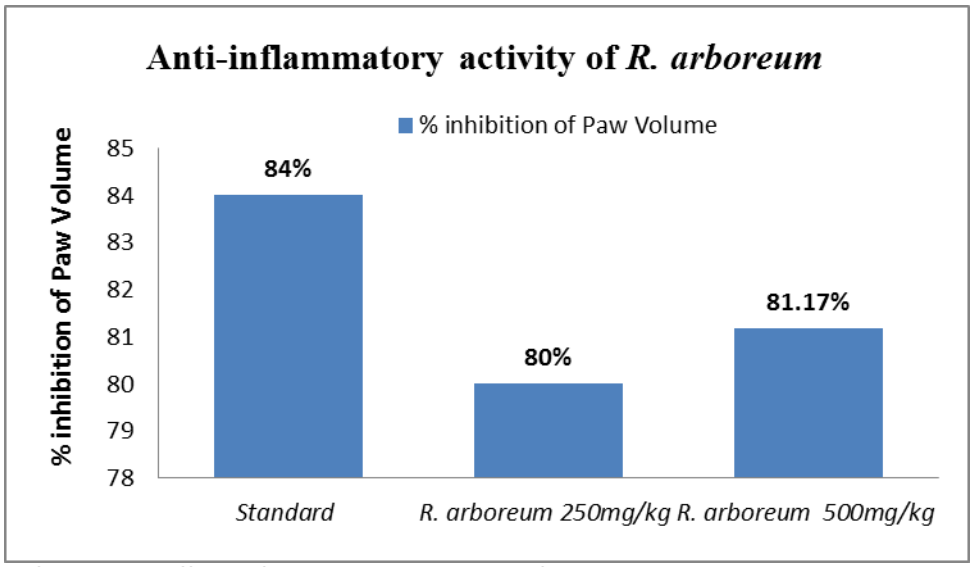

Fig 2: Percent of inhibition effects of the methanol extract of *Rhododendron arboreum* on Carrageenan induced paw edema in mice.

Results of Analgesic Activity Test

Table 4: Number of writhing and % of writhing inhibition.

Group	Number of Writhing (Mean ± SEM)	% of writhing inhibition
Control	41 ± 1.49	0
Standard	9 ± 0.70	78.04
R. arboretum 250 mg/kg	28 ± 1.08	31.70
R. arboretum 500 mg/kg	20 ± 1.79	51.21

Values were expressed in (Mean ± SEM) value. Each group comprised 4 animals. Control Group received normal water and standard group received 10mg/kg Diclofenac sodium. Extract group was treated with 250mg/kg and 500mg/kg of the crude extract of *Rhododendron arboreum*.

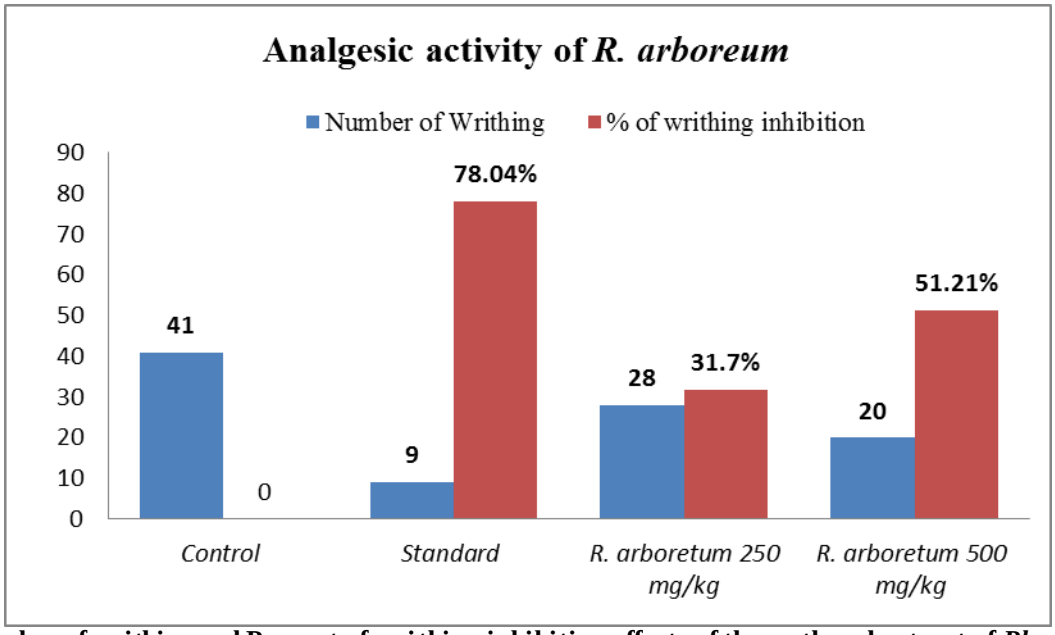

Fig 3: Number of writhing and Percent of writhing inhibition effects of the methanol extract of *Rhododendron arboreum* on analgesic activity test.

Results of Anti-diabetic Activity Test

Table 5: Plasma glucose level (mmol/L) of *Rhododendron arboreum leaves* extract in alloxan-induced diabetic (mM/L) in mice.

Animal Group	0min	30min	60min	90min	120min
Control Group	27.54±0.49	31.55±0.31	31.12±0.41	30.73±0.49	30.00±0.52
Standard Group	22.95±0.22	27.21±0.32	23.72±0.34	19.70±0.29	16.83±0.25
R. arboretum 250 mg/kg	26.83±0.20	28.67±0.25	26.32±0.35	25.19±0.19	24.43±0.23
R. arboretum 500 mg/kg	25.62±0.32	27.85±0.22	25.19±0.30	23.65±0.27	21.18±0.25

Values were expressed in (Mean ± SEM) value. Each group comprised 4 animals. Control Group received normal water and standard group received 100mg/kg Metformin Hydrochloride. Extract group was treated with 250mg/kg and 500mg/kg of the crude extract of *Rhododendron arboreum*.

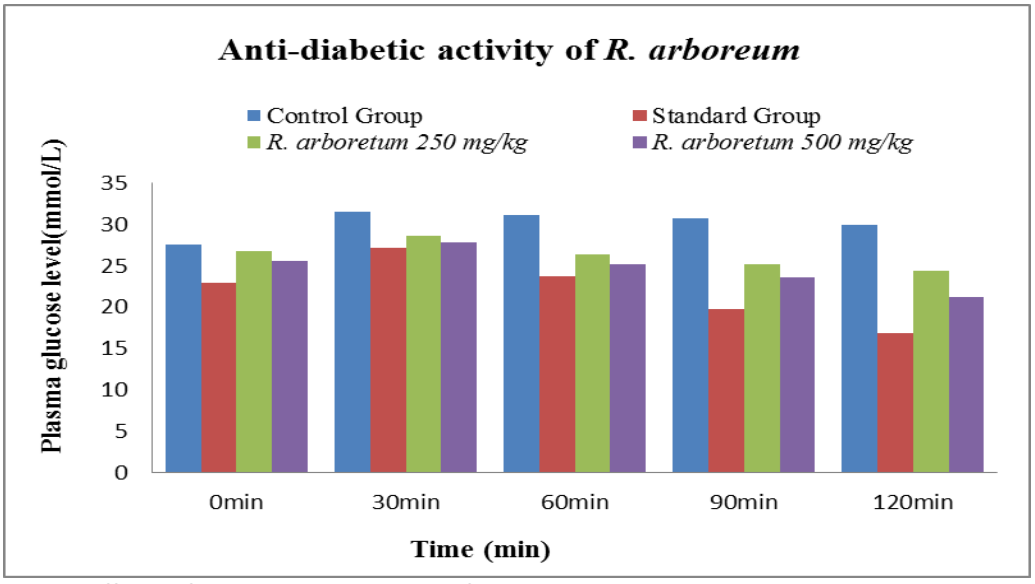

Fig 4: Effects of the methanol extract of *Rhododendron arboreum* on OGTT test in mice.

Table 6: % lessening of plasma glucose level.

Group	% lessening of plasma glucose level
Control	0
Standard	85.07
R. arboretum 250 mg/kg	63.44
R. arboretum 500 mg/kg	76.76

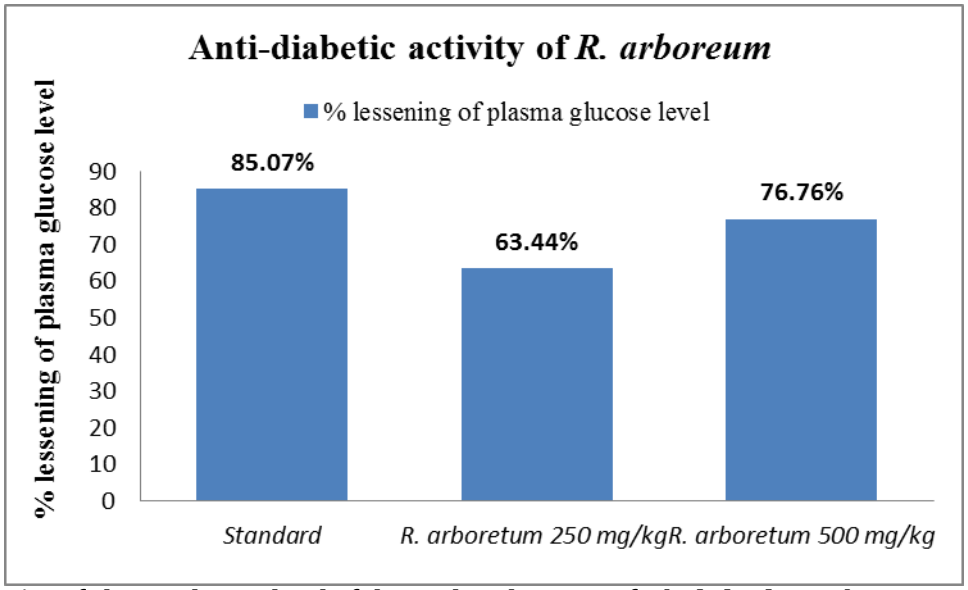

Fig 5: % lessening of plasma glucose level of the methanol extract of *Rhododendron arboreum* on OGTT test in mice.

DISCUSSION

Right now, pain relieving and mitigating exercises of methanol concentrate of Rhododendron arboretum leaves were surveyed in various very much acknowledged creature models, including Oral Glucose Tolerance Test (OGTT) in alloxan initiated diabetic mice, carrageenan actuated mice paw edema, acidic corrosive incited squirming test.

Diabetes mellitus is one of the most well-known ceaseless illnesses which raise the mortality fundamentally by type-2 diabetes. Unrecognized diabetes mellitus and hindered glucose resilience are generally connected with intense stroke.[38-40] The present investigation, concentrate of *Rhododendron arboretum* had set apart in hypoglycemic impacts in diabetic mice (Table-5 and Table-6).

Pain relieving drugs follows up on both CNS and PNS without altogether adjusting cognizance. The acidic corrosive actuated squirming is essentially a disclosure of fringe torment.[41-42] The acidic corrosive actuates arrival of various endogenous compound torment middle people like prostaglandin E2 (PG).[43-44] The analysis appeared (Table-4) that the concentrate displayed measurably huge hindrance of squirming at separate portion 250mg/kg and 500mg/kg body weight. The improvement of carrageenan-prompted edema is bi-phasic; the primary stage is credited to the arrival of histamine, serotonin and kinins and the subsequent stage is identified with the arrival of prostaglandins and bradykinins.[45-48] Mitigating detailed (Table-2 and Table-3) at portion 250mg/kg and 500mg/kg of methanol concentrate of *Rhododendron arboretum* leaves. It has been accounted for that various flavonoids have calming[49] and pain relieving[50] exercises. The nearness of flavonoid recognized may be liable for the pain relieving and calming exercises in methanol extricate.

CONCLUSION

The present assessment uncovered that concentrates of the *Rhododendron arboreum* leaves can be utilized as a wellspring of malignant growth avoidance specialist. Finally we can say that further evaluation is required to do in-vivo malignant growth avoidance specialist activity and locate the causative metabolites of *Rhododendron arboreum* leaves.

ACKNOWLEDGEMENT

I wish to offer significant thanks to Bangladesh University, Dhaka for their help to finish of this assessment adequately.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCE

1. Patel M, Murugananthan G, Gowda KP. In vivo animal models in preclinical evaluation of anti-inflammatory activity—A review. Int. J. Pharm. Res. Allied Sci., 2012; 1(2): 01-5.

2. Palladino MA, Bahjat FR, Theodorakis EA, Moldawer LL. Anti-TNF-α therapies: the next generation. Nature reviews Drug discovery. 2003 Sep; 2(9): 736-46.
3. Ferrero-Miliani L, Nielsen OH, Andersen PS, Girardin SE. Chronic inflammation: importance of NOD2 and NALP3 in interleukin-1β generation. Clinical & Experimental Immunology. 2007 Feb; 147(2): 227-35.
4. Pilotto A, Sancarlo D, Addante F, Scarcelli C, Franceschi M. Non-steroidal anti-inflammatory drug use in the elderly. Surgical Oncology. 2010 Sep 1; 19(3): 167-72.
5. Calixto JB, Campos MM, Otuki MF, Santos AR. Anti-inflammatory compounds of plant origin. Part II. Modulation of pro-inflammatory cytokines, chemokines and adhesion molecules. Planta medica. 2004 Feb; 70(02): 93-103.
6. Risso WE, Scarminio IS, Moreira EG. Antinociceptive and acute toxicity evaluation of Vernonia condensata Baker leaves extracted with different solvents and their mixtures.
7. Opdenakker G, Fibbe WE, Van Damme J. The molecular basis of leukocytosis. Immunology today. 1998 Apr 1; 19(4): 182-9.
8. Ricciotti E, FitzGerald GA. Prostaglandins and inflammation. Arteriosclerosis, thrombosis, and vascular biology. 2011 May; 31(5): 986-1000.
9. Grivennikov SI, Greten FR, Karin M. Immunity, inflammation, and cancer. Cell., 2010 Mar 19; 140(6): 883-99.
10. Moore MM, Chua W, Charles KA, Clarke SJ. Inflammation and cancer: causes and consequences. Clinical Pharmacology & Therapeutics. 2010 Apr; 87(4): 504-8.
11. Sosa S, Balick MJ, Arvigo R, Esposito RG, Pizza C, Altinier G, Tubaro A. Screening of the topical anti-inflammatory activity of some Central American plants. Journal of Ethnopharmacology. 2002 Jul 1; 81(2): 211-5.
12. Dassoler M, Schwanz M, Busseto F, Moreira EA, Gutierrez L. Perfil fitoquímico e ensaio farmacológico de Averrhoa carambola L.(Oxalidaceae). J Brasileiro Fitomedicina., 2004; 2: 4-8.
13. Lopes ER, Chapadeiro E, Raso P, Tafuri WL. Bogliolo Patologia, 1987: p. 67-112.
14. Ballantyne JC. Chronic pain following treatment for cancer: the role of opioids. The oncologist. 2003; 8(6): 567-75.
15. Fields HL. The doctor's dilemma: opiate analgesics and chronic pain. Neuron. 2011 Feb 24; 69(4): 591-4.
16. Gautam R, Jachak SM. Recent developments in anti-inflammatory natural products. Medicinal research reviews. 2009 Sep; 29(5): 767-820.
17. Besson JM. The neurobiology of pain. The Lancet. 1999 May 8; 353(9164): 1610-5.
18. McCurdy CR, Scully SS. Analgesic substances derived from natural products (natureceuticals). Life Sciences. 2005 Dec 22; 78(5): 476-84.
19. Nair SA, Shylesh BS, Gopakumar B, Subramoniam A. Anti-diabetes and hypoglycaemic properties of Hemionitis arifolia (Burm.) Moore in rats. Journal of ethnopharmacology. 2006 Jun 30; 106(2): 192-7.
20. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: estimates for the year 2000 and projections for 2030. Diabetes care. 2004 May 1; 27(5): 1047-53.
21. Malvi R, Jain S, Khatri S, Patel A, Mishra S. A review on antidiabetic medicinal plants and marketed herbal formulations. Int J Pharm Biol Arch., 2011; 2: 1344-55.
22. Safdar M, Khan A, Khattak MM, Siddique M. Effect of various doses of cinnamon on blood glucose in diabetic individuals. Pakistan Journal of Nutrition. 2004; 3(5): 268-72.
23. Ramachandran A, Das AK, Joshi SR, Yajnik CS, Shah S, Kumar KP. Current status of diabetes in India and need for novel therapeutic agents. J. Assoc. Physicians India. 2010 Jun; 58: 7-9.
24. Umar A, Ahmed QU, Muhammad BY, Dogarai BB, Soad SZ. Anti-hyperglycemic activity of the leaves of Tetracera scandens Linn. Merr.(Dilleniaceae) in alloxan induced diabetic rats. Journal of Ethnopharmacology., 2010 Aug 19; 131(1): 140-5.
25. Arulrayan N, Rangasamy S, James E, Pitchai D. A database for medicinal plants used in the treatment of diabetes and its secondary complications. Bioinformation., 2007 Jun; 2(1): 22-3.
26. Patel DK, Kumar R, Prasad SK, Sairam K, Hemalatha S. Antidiabetic and in vitro antioxidant potential of Hybanthus enneaspermus (Linn) F. Muell in streptozotocin–induced diabetic rats. Asian Pacific journal of tropical biomedicine., 2011 Aug 1; 1(4): 316-22.
27. Ponnusamy S, Ravindran R, Zinjarde S, Bhargava S, Ravi Kumar A. Evaluation of traditional Indian antidiabetic medicinal plants for human pancreatic amylase inhibitory effect in vitro. Evidence-Based Complementary and Alternative Medicine. 2011; 2011.
28. Li WL, Zheng HC, Bukuru J, De Kimpe N. Natural medicines used in the traditional Chinese medical system for therapy of diabetes mellitus. Journal of ethnopharmacology., 2004 May 1; 92(1): 1-21.
29. Saxena A, Vikram NK. Role of selected Indian plants in management of type 2 diabetes: a review. The Journal of Alternative & Complementary Medicine. 2004 Apr 1; 10(2): 369-78.
30. Jung M, Park M, Lee HC, Kang YH, Kang ES, Kim SK. Antidiabetic agents from medicinal plants. Current medicinal chemistry. 2006 Apr 1; 13(10): 1203-18.
31. Evans WC. Trease and Evans Pharmacognosy, International Edition E-Book. Elsevier Health Sciences; 2009 May 27.

32. Kannan R, Babu UV. Identity and pharmacognosy of Ruta graveolens Linn. Ancient science of life. 2012 Jul; 32(1): 16.
33. Shaheen SM. Hypoglycemic, Analgesic and Anti-Inflammatory Activities of Methanol Extract of Sida Rhombifolia L. Leaves on Experimental Mice. Int J Pharm Sci & Scient Res., 2017; 3: 7-82.
34. Winter CA, Risley EA, Nuss GW. Carrageenin-induced edema in hind paw of the rat as an assay for antiinflammatory drugs. Proceedings of the society for experimental biology and medicine. 1962 Dec; 111(3): 544-7.
35. Al-Haboubi HA, Zeitlin IJ. Re-appraisal of the role of histamine in carrageenan-induced paw oedema. European journal of pharmacology. 1983 Mar 25; 88(2-3): 169-76.
36. Ahmed F, Selim MS, Das AK, Choudhuri MS. Anti-inflammatory and antinociceptive activities of Lippia nodiflora Linn. Die Pharmazie-An International Journal of Pharmaceutical Sciences. 2004 Apr 1; 59(4): 329-30.
37. Hossain MS, Asadujjaman M, Khan MR, Ahmed M, Islam A. Antidiabetic and glycogenesis effects of different fractions of methanolic extract of Momordica charantia (Linn.) in alloxan induced diabetic rats. International Journal of Pharmaceutical Sciences and Research. 2011 Feb 1; 2(2): 404.
38. Cruickshank K. The epidemiology of diabetes mellitus.' Textbook of Diabetes. G Williams, J Pickup (eds). 1997: p. 17-23.
39. Haddad PS, Azar GA, Groom S, Boivin M. Natural health products, modulation of immune function and prevention of chronic diseases. Evidence-Based Complementary and Alternative Medicine. 2005; 2(4): 513-20.
40. Gray CS, Scott JF, French JM, Alberti KG, O'Connell JE. Prevalence and prediction of unrecognised diabetes mellitus and impaired glucose tolerance following acute stroke. Age and ageing. 2004 Jan 1; 33(1): 71-7.
41. Sanchez-Mateo CC, Bonkanka CX, Hernandez-Perez M, Rabanal RM. Evaluation of the analgesic and topical anti-inflammatory effects of Hypericum reflexum L. fil. Journal of ethnopharmacology. 2006 Aug 11; 107(1): 1-6.
42. Collier HO, Dinneen LC, Johnson CA, Schneider C. The abdominal constriction response and its suppression by analgesic drugs in the mouse. British journal of pharmacology and chemotherapy. 1968 Feb; 32(2): 295-310.
43. Zahid HI, Rizwani GH, Shareef HU, Ahmed MA, Hina BU. Analgesic and antipyretic activities of Hibiscus schizopetalus (Mast.) Hook. International Journal of Pharmacy and Pharmaceutical Sciences. 2012; 4(3): 218-21.
44. Deraedt R, Jouquey S, Delevallée F, Flahaut M. Release of prostaglandins E and F in an algogenic reaction and its inhibition. European journal of pharmacology. 1980 Jan 11; 61(1): 17-24.
45. Kapewangolo P, Omolo JJ, Bruwer R, Fonteh P, Meyer D. Antioxidant and anti-inflammatory activity of Ocimum labiatum extract and isolated labdane diterpenoid. Journal of Inflammation. 2015 Dec 1; 12(1): 4.
46. Dillingh MR, van Poelgeest EP, Malone KE, Kemper EM, Stroes ES, Moerland M, Burggraaf J. Characterization of inflammation and immune cell modulation induced by low-dose LPS administration to healthy volunteers. Journal of Inflammation. 2014 Dec 1; 11(1): 28.
47. Kavimani S, Mounissamy VM, Gunasegaran R. Analgesic and antiinflammatory activities of hispidulin isolated from Helichrysum bracteatum. INDIAN DRUGS-BOMBAY-. 2000; 37(12): 582-4.
48. Nivsarkar M, Mukherjee M, Patel M, Padh H, Bapu C. Launaea Nudicaulis leaf juice exhibits anti inflammatory action in acute and chronic inflammation models in rats. Indian drugs. 2002; 39(5): 290-2.
49. Sannigrahi S, Mazumder UK, Pal D, Mishra SL, Maity S. Flavonoids of Enhydra Fluctuans exhibits analgesic and anti-inflammatory activity in different animal models. Pakistan journal of pharmaceutical sciences. 2011 Jul 1; 24(3).
50. Hosseinzadeh H, Ramezani M, Fadishei M, Mahmoudi M. Antinociceptive, anti-inflammatory and acute toxicity effects of Zhumeria majdae extracts in mice and rats. Phytomedicine., 2002 Jan 1; 9(2): 135-41.