IJNRD.ORG ISSN : 2456-4184
INTERNATIONAL JOURNAL OF NOVEL RESEARCH AND DEVELOPMENT (IJNRD) | IJNRD.ORG
An International Open Access, Peer-reviewed, Refereed Journal

Therapeutic Efficacy of *Ficus religiosa* Leaves and *Punica granatum* Fruit

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Abstract

The use of medicinal plants for healing dates back as long as humanity. Since the beginning of time, medicinal plants have been utilized in healthcare. Worldwide research has been done to confirm their effectiveness, and some of the results have prompted the development of plant-based medications. Medicinal plant products have an annual market value of more than $100 billion worldwide. For millennia, researchers have researched inflammation with the goal of counteracting its effects on the body. Diabetes Mellitus is a metabolic condition affects around 10% of the global population, and the number of affected individuals rises daily. Globally, there is a growing trend toward the use of herbal treatments backed by in-depth laboratory study on the pharmacological properties of the bioactive compounds and their efficacy in treating a range of ailments. Studies on *Punica granatum* fruits and *Ficus religiosa* leaves have also been carried out recently to determine the presence of particular phytoconstituents. *Ficus religiosa* leaves and *Punica granatum* fruits have a variety of pharmacological properties, such as antioxidant, wound-healing, proteolytic, anticonvulsant, anti-diabetic, anti-inflammatory, and analgesic actions. Hence the objective of this present study is to determine the anti-inflammatory & anti-diabetic activity of the fresh and dried *Ficus religiosa* leaves & *Punica granatum* fruits. *In-vitro* anti-inflammatory and *in-vitro* anti-diabetic activity of the fresh and dried *Ficus religiosa* leaves & *Punica granatum* fruits were carried out by heat induced hemolysis & α-amylase test. Dried *Ficus religiosa* leaves and *Punica granatum* fruits have a better profile of anti-inflammatory & anti-diabetic activity compared to fresh *Ficus religiosa* leaves & *Punica granatum* fruits and thus it can be also used for lowering blood cholesterol levels, digestive problems & treating haematuria.

Keywords: Medicinal plants, Anti-Inflammatory, Anti-Diabetic, Pharmaceuticals, Phytochemicals.

I. Introduction

People have been searching for natural remedies to treat their illnesses since ancient times. As with animals, the usage of therapeutic herbs was initially instinctive (Stojanoski *et al.,* 1999). Medicinal plants are still a fascinating source of natural remedies for a range of medical ailments. An estimated 150,000 plant species have been researched; many of these include useful medicinal substances, and in recent years, there has been a progressive increase in the applications of novel chemicals from plants for pharmaceutical reasons (Shazhni *et al.,* 2018).

From ancient times, plants have been used extensively in human health treatment. Plants create several compounds with biological activity as a defense against pathogens and environmental stressors. These tiny chemical compounds have multiple biological functions and are produced by secondary metabolism. Anti-inflammatory properties are noted among the many uses (Virshette *et al.,* 2019).

It is well recognized that inflammation is a vital survival mechanism and a protective process that has been conserved throughout evolution. It is made up of intricately coordinated tissue changes aimed at eradicating the original source of the cell damage, which could have been physical (radiation, burns, trauma), chemical (caustic substances), or infectious agents or substances from their metabolism (microorganisms and toxins) (Fialho *et al.,* 2018).

Many phytoconstituents having antidiabetic properties, such as terpenoids, saponins, flavonoids, carotenoids, alkaloids, and glycosides, are found in medicinal plants. Numerous phytoconstituents are carried by the intricate plant matrix, which controls how these compounds interact. This is beneficial to health but is also difficult to replicate (Durazzo *et al.,* 2018).

Diabetes mellitus is fast-moving. 9.3% of adults worldwide were determined to have diabetes in 2019. 352 million individuals were at risk of type 2 diabetes in 2017, according to a research by the International Diabetes Federation. By 2030, 439 million persons are expected to develop diabetes, according to health projections. Diabetes increased mortality in individuals with COVID-19 in a meta-analysis of studies done during the pandemic. In 2010, diabetes accounted for 5.7% of all deaths in North America and 6% of all deaths in Africa (Wu *et al.,* 2021).

Generally speaking, equilibrium is restored as a result of this intricate biological reaction. However, the inflammatory process continues and a modest but persistent pro-inflammatory state may emerge in situations of prolonged release of inflammatory mediators and the activation of detrimental signal-transduction pathways (Liu *et al.,* 2017). Numerous illnesses and long-term medical issues, including obesity, diabetes, cancer, and cardiovascular diseases, are associated with a low-grade inflammatory state (Kim *et al.,* 2018).

The presence of several advantageous chemical components in *Ficus religiosa* leaves has drawn the attention of the medical community. *Ficus religiosa* leaves are widely used to cure a variety of ailments, including

diabetes, inflammation, respiratory conditions, gastrointestinal disorders, sexual dysfunction, and abnormalities of the central nervous system (CNS), according to a number of research on the subject (Priya et al., 2020).

These days, *Punica granatum* are reported to have therapeutic benefits for a wide range of illnesses and conditions, such as obesity, diabetes, aging, and inflammation, in international literature. In particular, it has been demonstrated that treating obesity using *Punica granatum* -derived natural chemicals is effective (Viladomiu *et al.*, 2013).

Thus, the goal of this research is to determine the amount of anti- inflammatory and anti-diabetic activity in both fresh and dried *Ficus religiosa* leaves and *Punica granatum* fruits. Hence the present study is carried out by following objectives are to determine the *in-vitro* anti-inflammatory activity & *in-vitro* anti-diabetic activity of fresh and dried *Ficus religiosa* leaves and *Punica granatum* fruits.

II. Materials and Methods

2.1 Selection and collection of plant materials

The *Ficus religiosa* leaves are serve as an expectorant, diuretic, ointment. The juice of these leaves brings down nausea, cleanses digestive system and maintains skin health. Being strongly astringent in properties, *Ficus religiosa* leaves when heated exudes purgative properties and treated for digestive problem. *Punica granatum* fruits can have up to three times more antioxidants than green tea or red wine. They provide plenty of macro- and micronutrients, as well as bioactive compounds that promote health. They also have anti-diabetic, anti-carcinogenic activity and improve urinary health. Hence these ingredients are selected for the present study for due to the presence of Anti-inflammatory and Anti-diabetic activity. The *Ficus religiosa* leaves and *Punica granatum* fruits were collected from Murugampalayam, Tirupur during September 2023. It was authenticated by **Scientist 'F' Dr. K.Karthigeyan** Botanical survey of India, Coimbatore and the letter No. BSI/SRC/5/23/2023/Tech-177 for *Ficus religiosa* and letter No. BSI/SRC/5/23/2023/Tech-178 for *Punica granatum*.

2.2 Processing of Fresh and Dried *Ficus religiosa* leaves:

The sample was first visually examined for any kind of infection, spores, damage, discoloration, and distortion. Undamaged samples of leaf were thoroughly washed with tap water, then rinsed using distilled water. The midribs of leaves were removed. The dried leaves are shadow dried for five days and then grinded into fine powder and stored in airtight bottles. Natural shade drying is the most accepted storage method for aromatic medicinal herbs because of its low cost and minimum loss of volatile constituents.

2.3 Processing of Fresh and Dried *Punica granatum* fruits:

The sample was first visually examined for any kind of infection, spores, damage, discoloration, and distortion. Undamaged samples of fruits were collected, grinded and filter to get extract. Natural shade drying is the most accepted storage method for aromatic medicinal herbs because of its low cost and minimum loss of volatile constituents. So the fruits are shadow dried for 3-4 days, collected & then grinded into a fine powder and stored in an air tight container.

2.4 Extraction Process – Maceration

Maceration is the process of placing coarsely crushed drug material—such as leaves, stem bark, or root bark—into a container and pouring menstrual fluid over it until the drug material is completely covered. Following that, the container is sealed and left for at least three days. If the material is stored in a bottle, it is shaken and mixed on a regular basis to ensure complete extraction. After extraction, the micelle and mark are separated via decantation or filtration. The micelle is next evaporated in an oven or over a water bath to separate it from the menstruum. This method is practical and suitable for materials that are thermo stable in plants (Ingle *et al.*, 2017).

The extraction was done with aqueous solvent for *in-vitro* anti-inflammatory Activity & *in-vitro* anti-diabetic activity.

2.5 *In-Vitro* Anti-Inflammatory Activity

Heat-Induced Hemolysis

The method had been previously described by Shinde *et al*. (1999) and slightly modified and followed by Henneh *et al.* (2018). The reaction mixture (2 ml) consisted of 1.0 ml of 10% HRBC and 1 ml of various solvents plant extracts (1 mg/ml), which was added to each tube and gently mixed. The positive control consisted of 1.0 ml of HRBC and 1.0 ml of various concentrations of diclofenac sodium (10 to 50 µg/ml). The negative control consisted of 1.0 ml of 10% erythrocyte suspension and 1.0 ml of normal saline alone. The experiment was performed in triplicates. The resulting solution was heated at 56° C for 30 minutes and cooled to room temperature and centrifuged at 2500 rpm for 10 minutes. The supernatant was collected and the absorbance of each solution was measured spectrophotometrically (UVmini 1240, Shimadzu) at 560 nm as an indicator of the degree of haemolysis. The percentage inhibition of hemolysis was calculated using the formula

$$\text{Percentage of inhibition} = \frac{Ac - At}{Ac} \times 100$$

Where ‘Ac’ is absorbance of control and ‘At’ is absorbance of the test.

2.6 *In-Vitro* Anti-Diabetic Activity

Inhibition assay for α-amylase activity

α-amylase was premixed with extract at various concentrations (50-250 µg/mL) and starch as a substrate was added (0.5% starch solution) to start the reaction. The reaction was carried out at 37°C for 5 min and terminated by addition of 2 mL of DNS (3,5-dinitrosalicylic acid) reagent. The reaction mixture was heated for 15 min at 100°C and diluted with 10 mL of distilled water in an ice bath (Miller, 1959). α-amylase activity was determined by measuring spectrum at 540 nm. The IC_{50} value was defined as the concentration of α-amylase inhibitor to inhibit 50% of its activity under the assay conditions. The percentage inhibition assay for a α-amylase activity was calculated using the formula

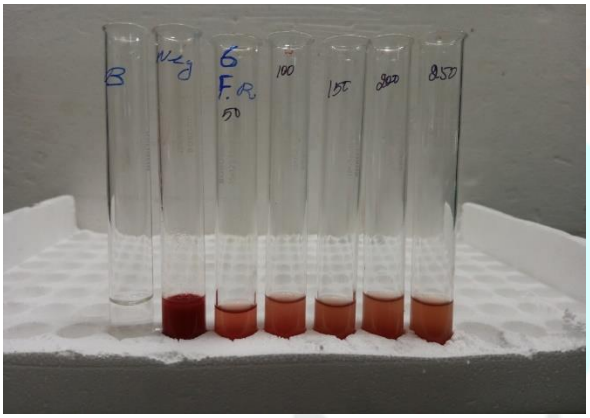
$$\text{Percentage of inhibition} = \frac{Ac - At}{Ac} \times 100$$

Where 'Ac' is absorbance of control and 'At' is absorbance of the test.

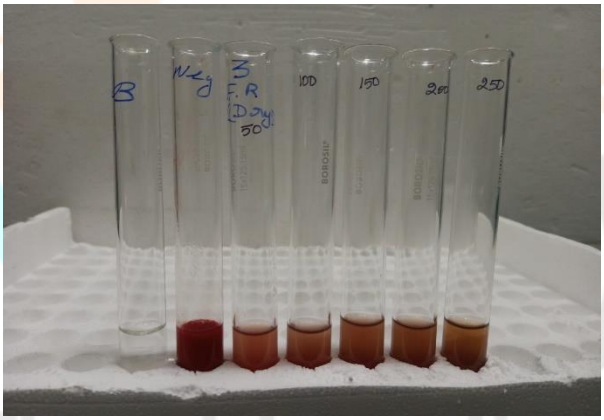
III. Results

3.1 *In-Vitro* Anti-Inflammatory Activity

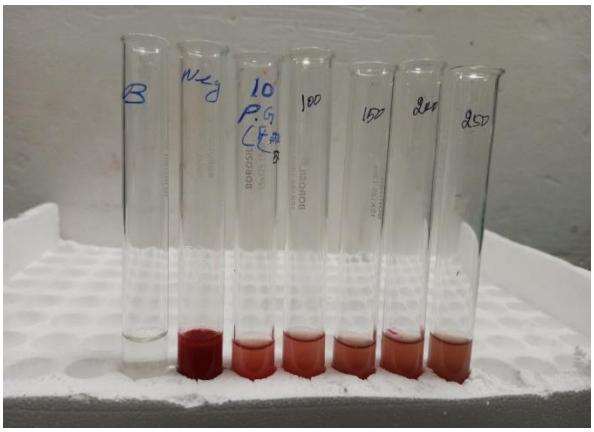
Heat-Induced Hemolysis



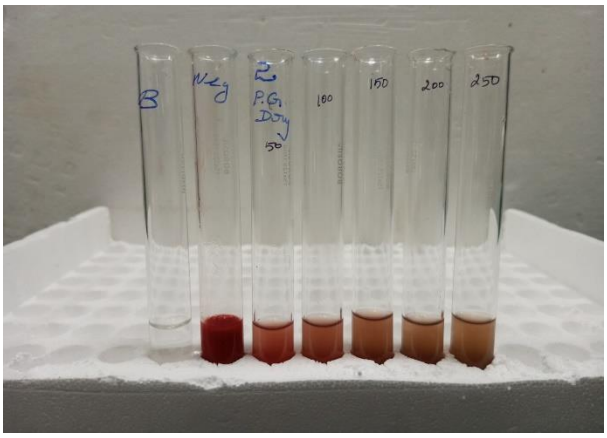
Fresh *Ficus religiosa* leaves



Dried *Ficus religiosa* leaves



Fresh *Punica granatum* fruit



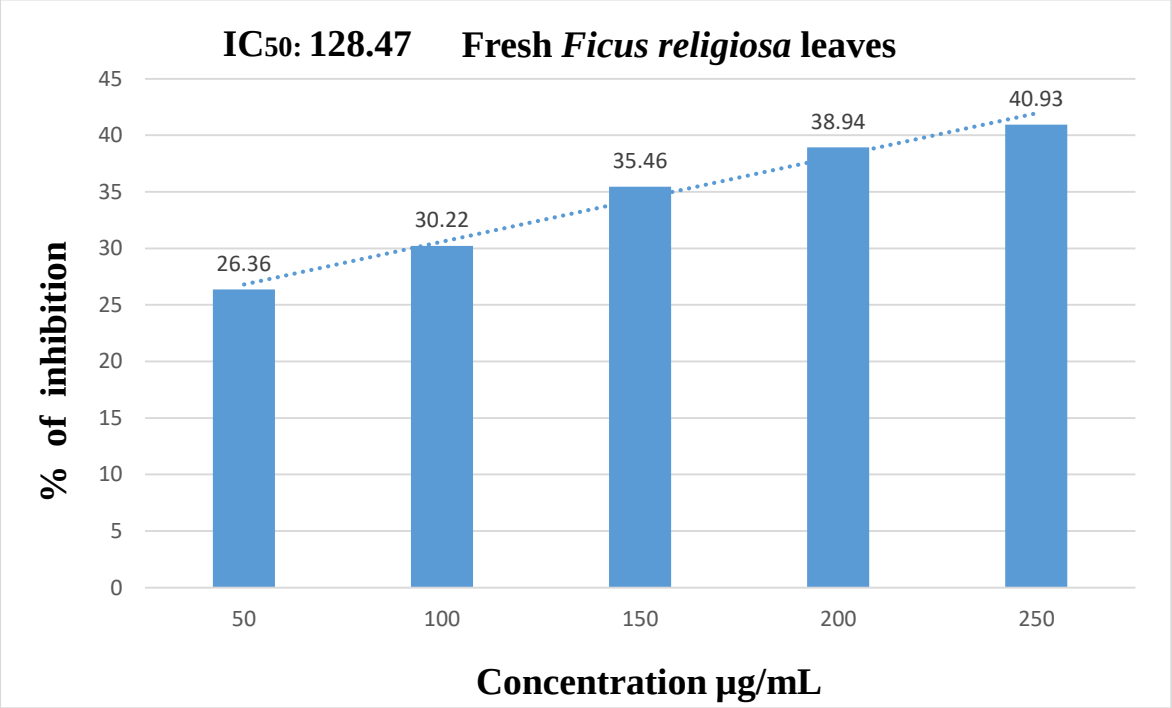
Dried *Punica granatum* fruit

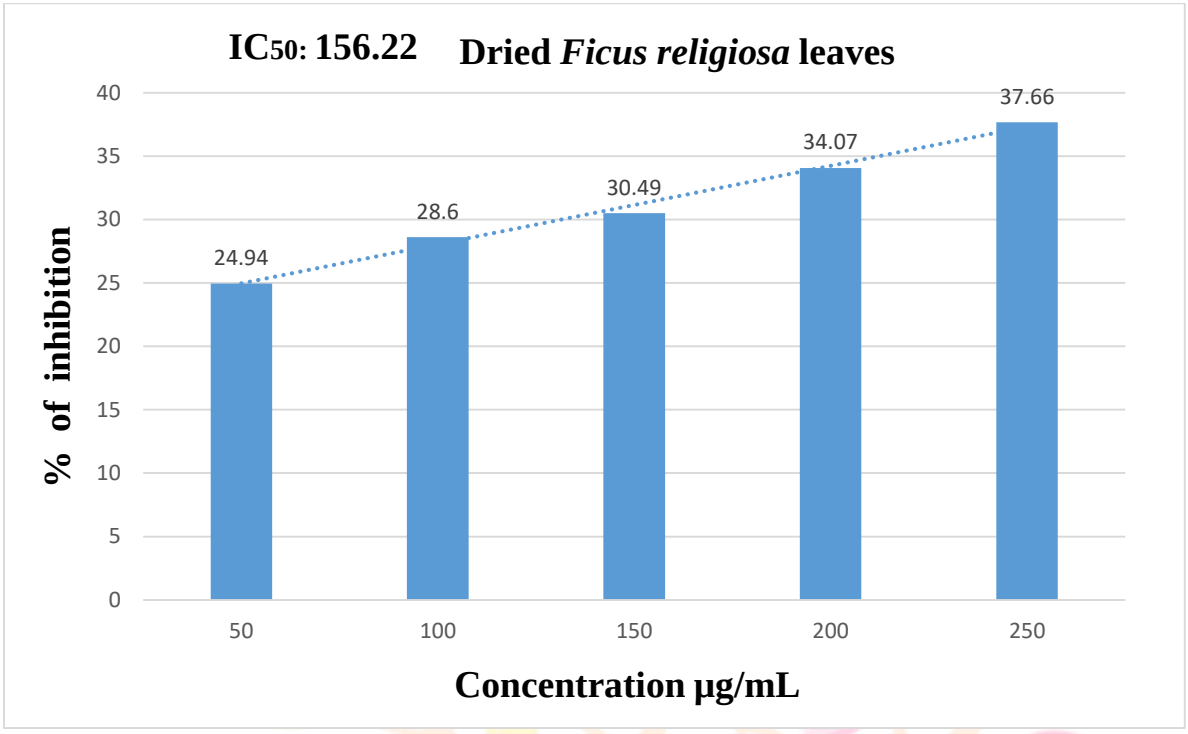
Table No: 3.1 Heat-Induced Hemolysis

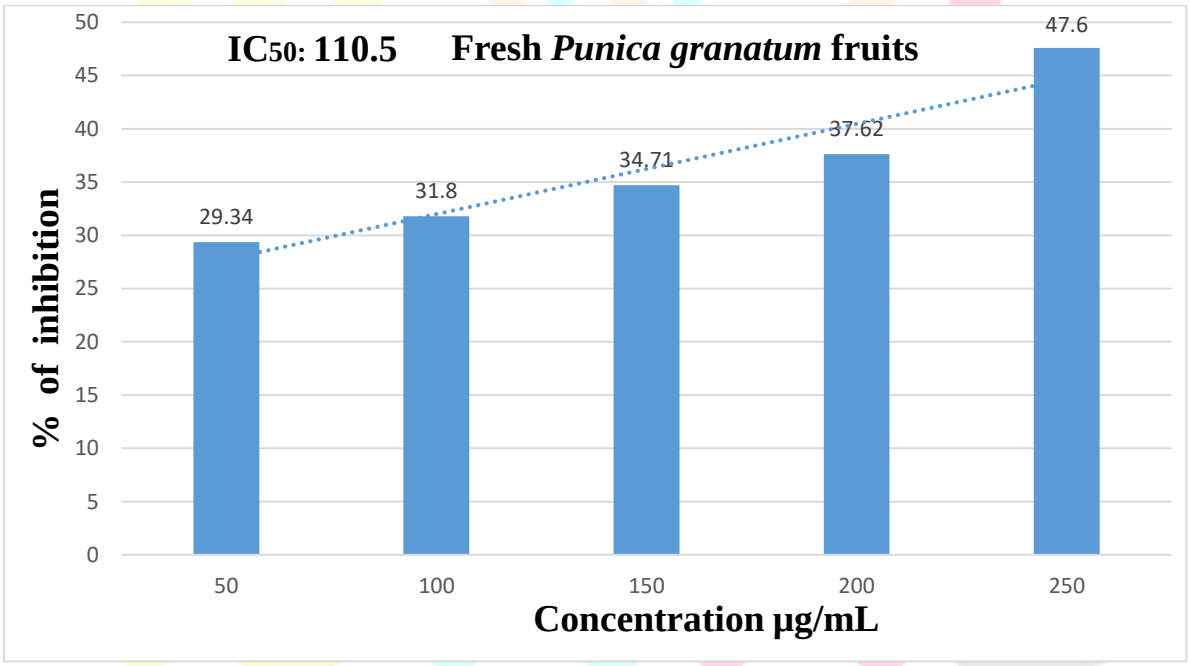
Extracts	% of Inhibition		% Scavenging activity IC_{50} (µg/mL)
Ficus religiosa leaves	50µl	26.36	128.47
	100µl	30.22	
	150µl	35.46	
	200µl	38.94	
	250µl	40.93	
Dried *Ficus religiosa* leaves	50µl	24.94	156.22
	100µl	28.60	
	150µl	30.49	
	200µl	34.07	
	250µl	37.66	
Punica granatum fruits	50µl	29.34	110.5
	100µl	31.87	
	150µl	34.71	
	200µl	37.62	
	250µl	47.76	
	50µl	27.28	215.17
	100µl	29.85	

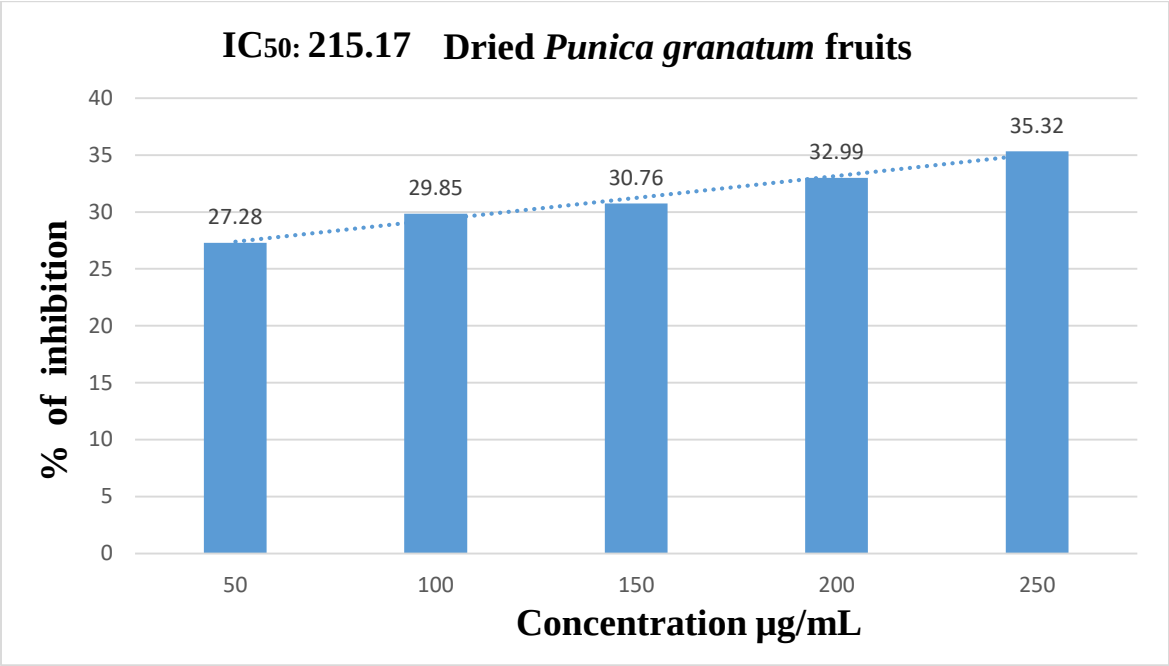
Dried *Punica granatum fruits*	150µl	30.76	
	200µl	32.99	
	250µl	35.32	

Table No: 4.1 depicts the anti-inflammatory activity of fresh and dried *Ficus religiosa* leaves and *Punica graantum* fruits. Heat induced hemolysis activity in fresh *Ficus religiosa* leaves (26.36%, 30.22%, 35.46%, 38.94%, 40.93%) & fresh *Punica granatum* fruit (29.34%, 31.87%, 34.71%, 37.62%, 47.76%) contain more anti-inflammatory activity than dried *Ficus religiosa* leaves (24.94%, 28.60%, 30.49%, 34.07%, 37.66%) & dried *Punica granatum* fruit (27.28%, 29.85%, 30.76%, 32.99%, 35.32%).



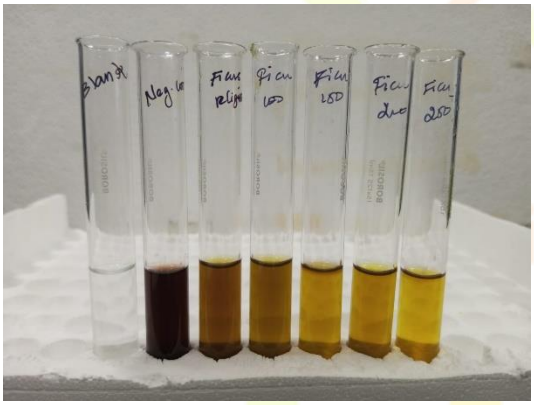




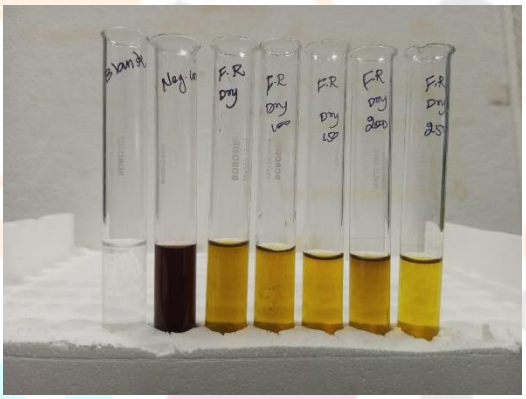


3.2 *In-vitro* Anti-Diabetic Activity

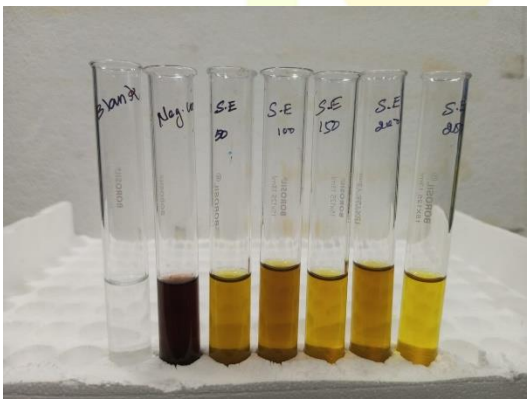
Inhibition assay for α-amylase activity



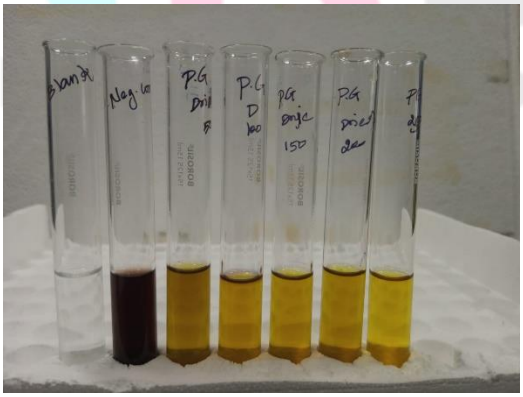
Fresh *Ficus religiosa* leaves



Dried *Ficus religiosa* leaves



Fresh *Punica granatum* fruit

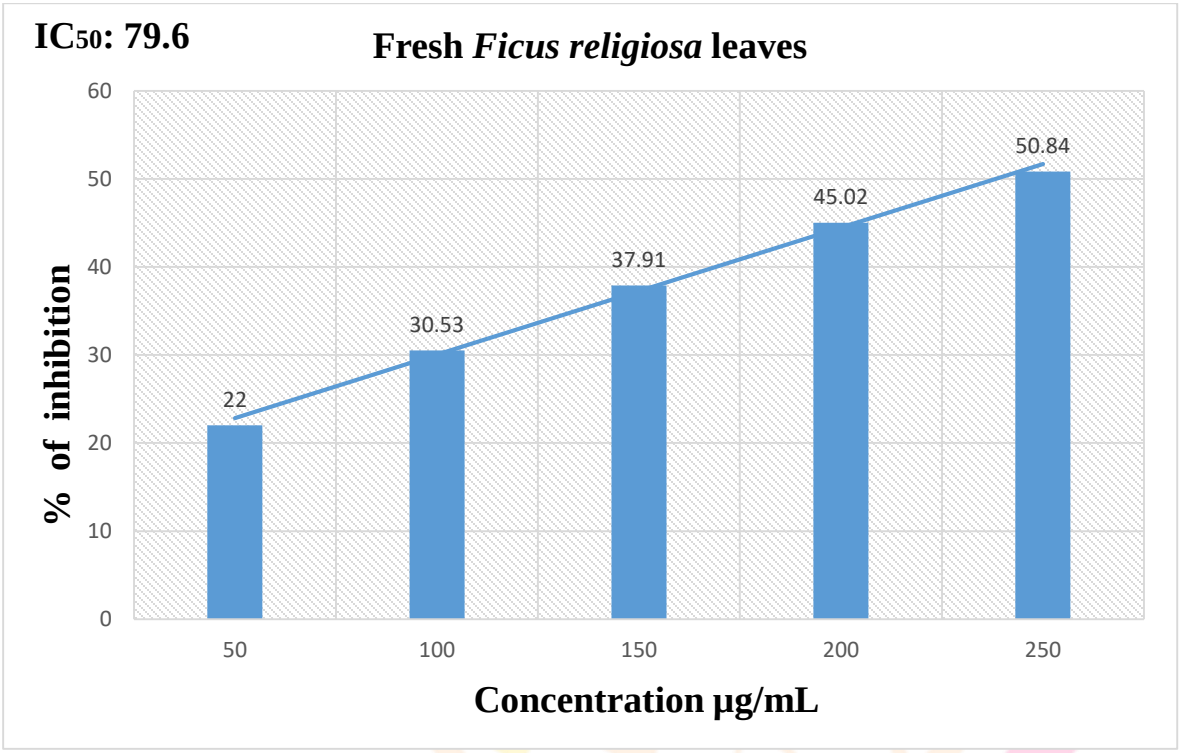


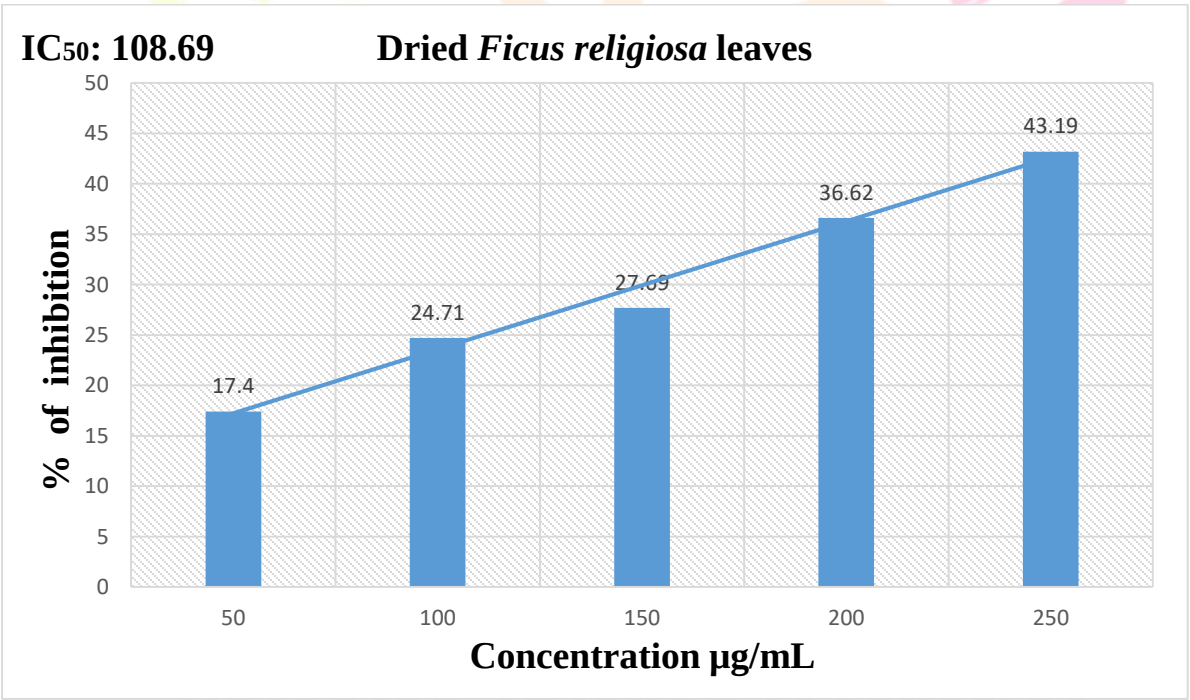
Dried *Punica granatum* fruit

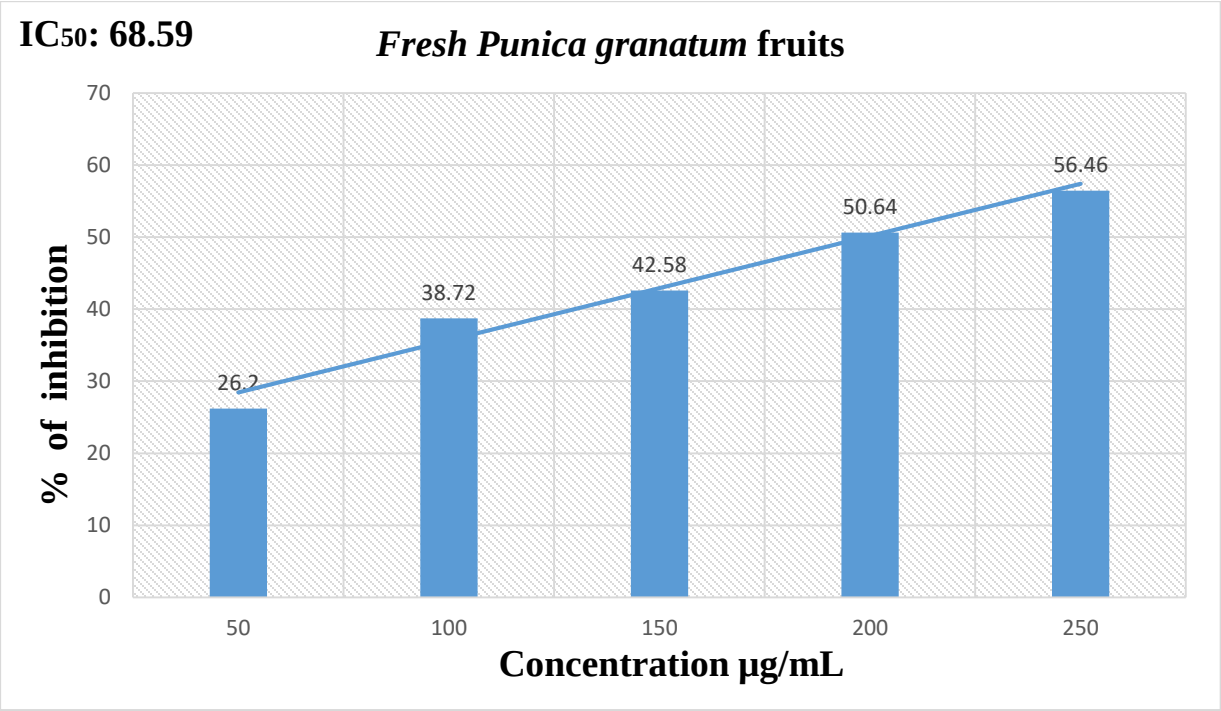
Table No: 4.2 Inhibition assay for α-amylase activity

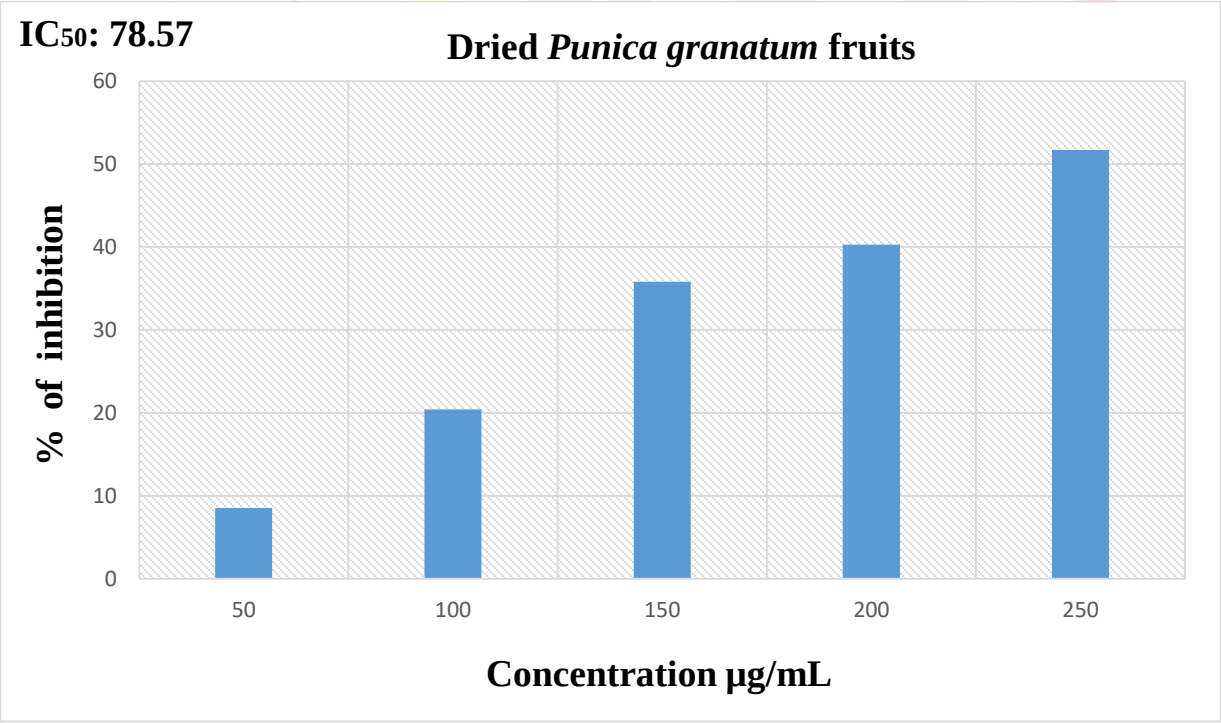
Extracts		% of Inhibition	% Scavenging activity IC_{50} (µg/mL)
Ficus religiosa leaves	50µl	22	79.60
	100µl	30.53	
	150µl	37.91	
	200µl	45.02	
	250µl	50.84	
Dried *Ficus religiosa* leaves	50µl	17.40	108.69
	100µl	24.71	
	150µl	27.69	
	200µl	36.62	
	250µl	43.19	
Punica granatum fruits	50µl	26.20	68.59
	100µl	38.72	
	150µl	42.58	
	200µl	50.64	
	250µl	56.46	
Dried *Punica granatum fruits*	50µl	8.53	78.57
	100µl	20.44	
	150µl	35.81	
	200µl	40.28	
	250µl	51.72	

Table No: 4.2 depicts the anti-diabetic activity of fresh and dried *Ficus religiosa* leaves and *Punica graantum* fruits. Inhibition assay for α-amylase activity in fresh *Ficus religiosa* leaves (22%, 30.53%, 37.91%, 45.02%, 50.84%) & fresh *Punica granatum* fruit (26.20%, 38.72%, 42.58%, 50.64%, 56.46%) contain more anti-diabetic activity than dried *Ficus religiosa* leaves (17.40%, 24.71%, 27.69%, 36.62%, 43.19%) & dried *Punica granatum* fruit (8.53%, 20.44%, 35.81%, 40.28%, 51.72%).


IC50: 79.6
Fresh Ficus religiosa leaves
% of inhibition
60
50
40
30
20
10
0
22
30.53
37.91
45.02
50.84
50
100
150
200
250
Concentration µg/mL


IC50: 108.69
Dried Ficus religiosa leaves
% of inhibition
50
45
40
35
30
25
20
15
10
5
0
17.4
24.71
27.69
36.62
43.19
50
100
150
200
250
Concentration µg/mL





IV. Discussion

There are many methods to estimate the anti-inflammatory action of drugs and anti-diabetic activity. The extract was effective in reducing the heat-induced hemolysis at various concentrations. The results showed that maximum inhibition was with aqueous extract of fresh *Ficus religiosa* leaves with 40.93% at 250µl/ml compared to dried *Ficus religiosa* leaves with 37.66% at 250µl/ml while in fresh *Punica granatum* fruits with 47.76% at

250µl/ml has maximum inhibition compared to dried *Punica granatum* fruits with 35.32% at 250µl/ml. The results showed that the anti-diabetic effectively inhibited the α-amylase enzyme activity with a maximum inhibition of fresh *Ficus religiosa* leaves with 50.84% at a concentration of 250µg/ml compared to dried Ficus religiosa leaves with 43.19% while in fresh *Punica granatum* fruit has maximum inhibition of 56.46% at the concentration of 250µl/ml compared to dried *Punica granatum* fruit with 51.72% at 250µl/ml.

V. Conclusion

The most valuable and well-known plants in the Ayurvedic medicine are the pomegranate (*Punica granatum*) and peepal (*Ficus religiosa*). They are among the most versatile plants and have many different medicinal applications. Phytochemicals, minerals, and antioxidants provide leaves & fruits unique therapeutic qualities. Rich in vitamins and minerals, the leaves of *Ficus religiosa* trees and the fruits of *Punica granatum* are powerful antioxidants. They improve digestion and oral health, boost renal function, reduce blood sugar, have anti-cancer, anti-heart disease, and wound-healing effects. The present study revealed that the aqueous extract of fresh *Ficus religiosa* leaves & *Punica granatum* fruits showed the highest of anti-inflammatory & anti-diabetic activity.

References

1. Stojanoski N. (1999). "Development of health culture in Veles and its region from the past to the end of the 20th century." *Veles: Society of science and ar*, 13–34.

2. Shazhni J.A, Renu A, Vijayaraghavan P. (2018). "Insights of antidiabetic, anti-inflammatory and hepatoprotective properties of antimicrobial secondary metabolites of corm extract from Caladium x hortulanum." *Saudi J. Boil. Sci*, *25*, 1755–1761.

3. Locatelli C, Nardi G.M, Anuário A.d.F, Freire C.G, Megiolaro F, Schneider K, Perazzoli M.R.A, Nascimento S.R.D, Gon A.C, Mariano L.N.B. (2016). "Anti-inflammatory activity of berry fruits in mice model of inflammation is based on oxidative stress modulation." *Pharmacogn. Res, 8*, S42–S49.

4. Virshette S.J, Patil M.K, Somkuwar A.P. (2019). "A review on medicinal plants used as anti-inflammatory agents." *J. Pharmacogn. Phytochem*, *8*, 1641–1646.

5. Fialho L, Cunha-E-Silva J.A, Santa-Maria A.F, Madureira F.A, Iglesias A.C. (2018). "Comparative study of systemic early postoperative inflammatory response among elderly and non-elderly patients undergoing laparoscopic cholecystectomy." *Rev. Col. Bras. Cir*, *45*, e1586.

6. Jang C.H, Kim Y.Y, Seong J.Y, Kang S.H, Jung E.K, Sung C.M, Kim S.B, Cho Y.B, Sung J.Y. (2016). "Clinical characteristics of pediatric external auditory canal cholesteatoma." *Int. J. Pediatr. Otorhinolaryngol*, *87*, 5–10.

7. Liu C.H, Abrams N, Carrick D.M, Chander P, Dwyer J, Hamlet, M.R.J, Macchiarini F, Prabhudas M, Shen G.L, Tandon P, et al. (2017). "Biomarkers of chronic inflammation in disease development and prevention: Challenges and opportunities." *Nat. Immunol*, *18*, 1175–1180.

8. Kim Y, Bayona P.W, Kim M, Chang J, Hong S, Park Y, Budiman A, Kim Y.-J, Choi C.Y, Kim W.S, et al. (2018). "Macrophage Lamin A/C Regulates Inflammation and the Development of Obesity-Induced Insulin Resistance." *Front. Immunol*, *9*, 1–14.

9. Purohit S, Sharma A, Zhi W, Bai S, Hopkins D, Steed L, Bode B, Anderson S.W, Reed J.C, Steed R.D, et al. (2018) "Proteins of TNF-α and IL6 Pathways Are Elevated in Serum of Type-1 Diabetes Patients with Microalbuminuria." *Front. Immuno, 9,* 154.

10. Durazzo A, D'Addezio L, Camilli E, Piccinelli R, Turrini A, Marletta L, Marconi S, Lucarini, Lisciani S, Gabrielli P, et al. (2018). "From plant compounds to botanicals and back: A current snapshot." *Molecules,* 23:1844. doi: 10.3390/molecules23081844.

11. Wu Z, Tang Y, Cheng Q. (2021). "Diabetes increases the mortality of patients with COVID-19: A meta-analysis." *Acta Diabetol,*58:139–144. doi: 10.1007/s00592-020-01546-0.

12. Viladomiu M, Hontecillas R, Lu P, Bassaganya-Riera J. (2013). "Preventive and prophylactic mechanisms of action of pomegranate bioactive constituents." *Evid. Based Complement. Altern. Med,* 789764.

13. Tiwari Priya, Gupta Rishikesh. (2020). "Preliminary phytochemical screening of bark (powder) extracts of *Ficus religiosa* (Peepal) Plant". *International Journal of Research and Development in Pharmacy & Life Science,* 9(1): 1-6.

14. Ingle KP, Deshmukh AG, Padole DA, Dudhare MS, Moharil MP, Khelurkar VC. (2017). "Phytochemicals: Extraction methods, identification, and detection of bioactive compounds from plant extracts." *J Pharmacogn Phytochem,* 6:32–6.