

# Pharmacological Investigations of Ethanolic Roots Extract of *Asparagus racemosus* Willd

<sup>1</sup>Pratibha Singh, <sup>2</sup>Amrita Singh, <sup>3</sup>Muskan Saxena, <sup>4</sup>Bhanu P. S. Sagar

IEC Department of Pharmacy  
IEC Group of Institutions  
Greater Noida, Uttar Pradesh, India

**Abstract-** Identity and purity of shatavari roots were established pharmacognostically and herbarium specimen IEC/Pharm/Herb/2022/2224 was deposited in Herbarium Bank. Shatavari roots confirmed phenolic compounds, alkaloids, sterols, flavanoids, steroidal glycosides isoflavones, and tannins. In physicochemical analysis of roots of *Asparagus racemosus*, roots were free from contamination (low ash values), AIAV 1.6 % (Low acid-insoluble ash values) indicated drug was free from siliceous matters / sand, WSAV 4.42 % (Watersoluble ash value) indicated presence of water soluble salts and ASEV (7.94 %), WSEV (8.62%) (solvent extractive values) indicated presence of phyto-constituents and 2.68% (LOD). *Asparagus racemosus* Willd. EERAR extract induced powerful Rat RBC membrane stabilizing property (45.24% to 86.56% membrane stabilization impact of EERAR). *Asparagus racemosus* Willd. (shatavari) EERAR (A/B) produced impactful anti-inflammatory activity. The indomethacin induced excellent anti-inflammatory (edema reduced significantly). *Asparagus racemosus* Willd. EERAR roots extract contains phenolics, sterols and flavonoids possess significant inflammatory effects. Indomethacin and EERAR-B anti-inflammatory effects were very good in comparison of drug vehicle control group (significant at  $P < 0.05$  over control). Further, in Cotton pellet-induced granuloma method, Indomethacin (10 mg/kg) inhibition 53.20% ; EERAR-B: (400 mg/kg) inhibition 32.82%; values were significant at  $p < 0.05$ ).

**Keywords:** Anti-inflammatory, carageenan, flavonoids, herbal medicines, indomethacin, natural products. paw edema, physicochemical, shatavari, solvent extractive values,

## General Introduction

### Inflammation

Campos *et al.*, 2014, Inflammation is a ubiquitous (disturb homeostasis) and severe cause of morbidity. Burke *et al.*, 2005, Inflammation is a natural aspect of the immune system's response. In Inflammation (body's severe reaction to any damage), pain, redness, heat or warmth, and swelling are the four primary indicators of swelling and in injury arterioles in the local tissue dilate and results in increased blood flow to the affected area (redness).

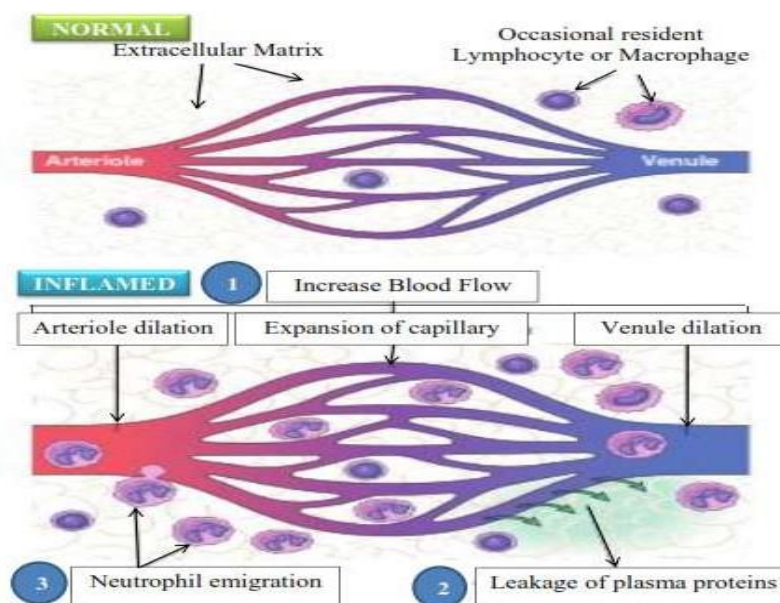


Figure 1 : Inflammatory Process.

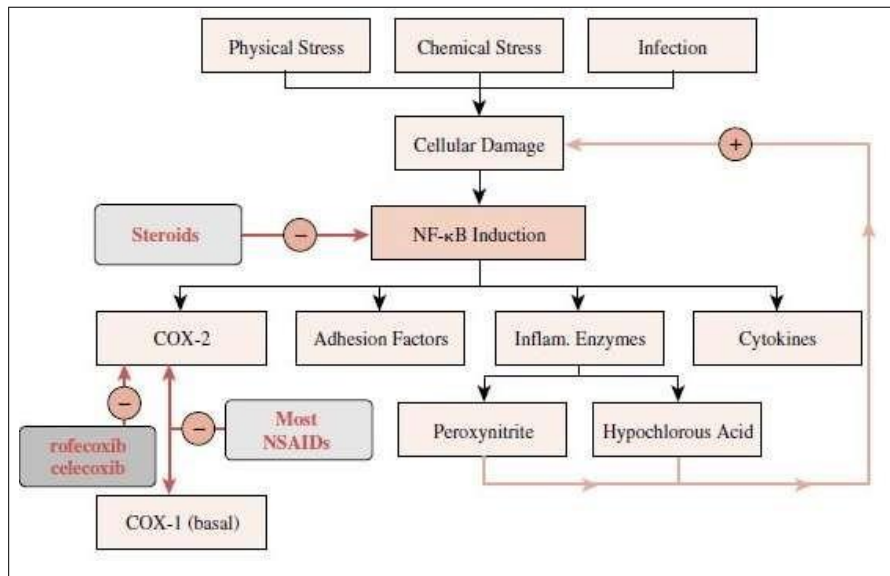


Figure 2 : Overview of Inflammatory Processes.

**Signs of Inflammation**

Parhnam *et al.*, 2008, signs of inflammation include redness (local), swelling, pain, heat and loss of function (chemical mediators, including kinins, eicosanoids, complement proteins, histamine and monokines induce and regulate these manifestations).



Figure 3 : Signs of inflammation.

**Types of Inflammation**

Libby *et al.*, 2003, inflammation is either acute or chronic. Acute inflammation is an initial response of the body to harmful stimuli. In chronic inflammation, the response resulting in damage to the body (out of proportion; rheumatoid arthritis, asthma, colitis, allergies, hepatitis, metabolic syndrome, autoimmune diseases cancer, cardiovascular dysfunctions and neurodegenerative disorders).

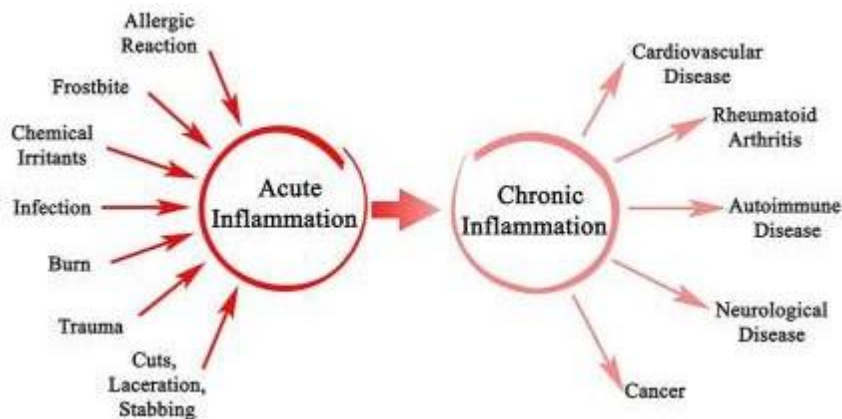
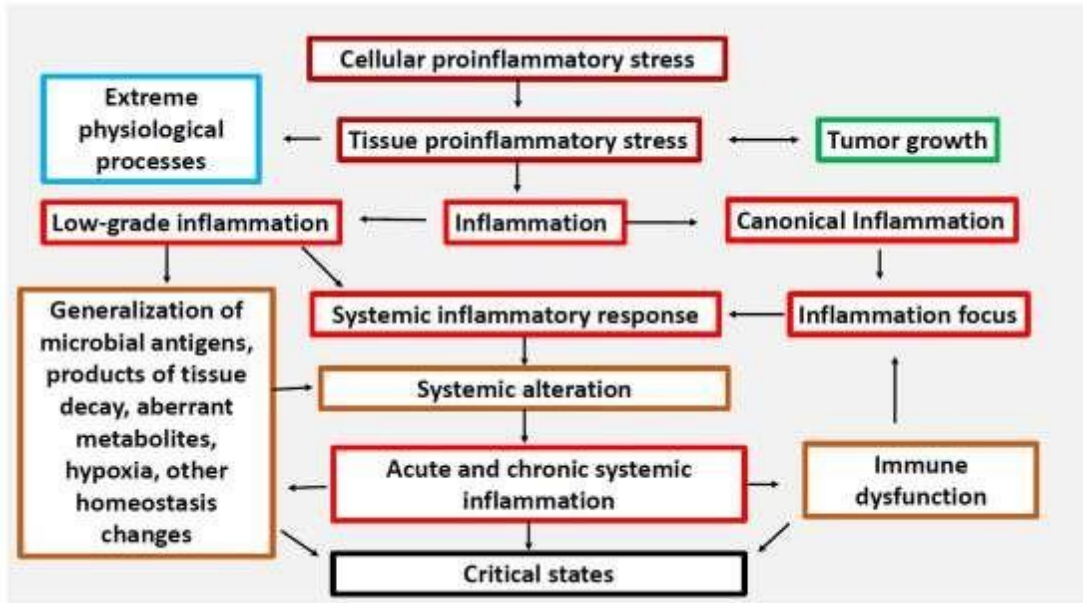


Figure 4: Types of Inflammation.

Pathophysiology of Inflammation



Note: The Inflammatory Cascade is a non-specific response to all foreign pathogens and trauma.

Abbreviations:  
ILs: Interleukins  
WBC's: White Blood Cells

reviewer  
Meghan Jacksc  
Sean Dohert  
Dr. Luis Murgula Favela  
\* MD at time of publicatio

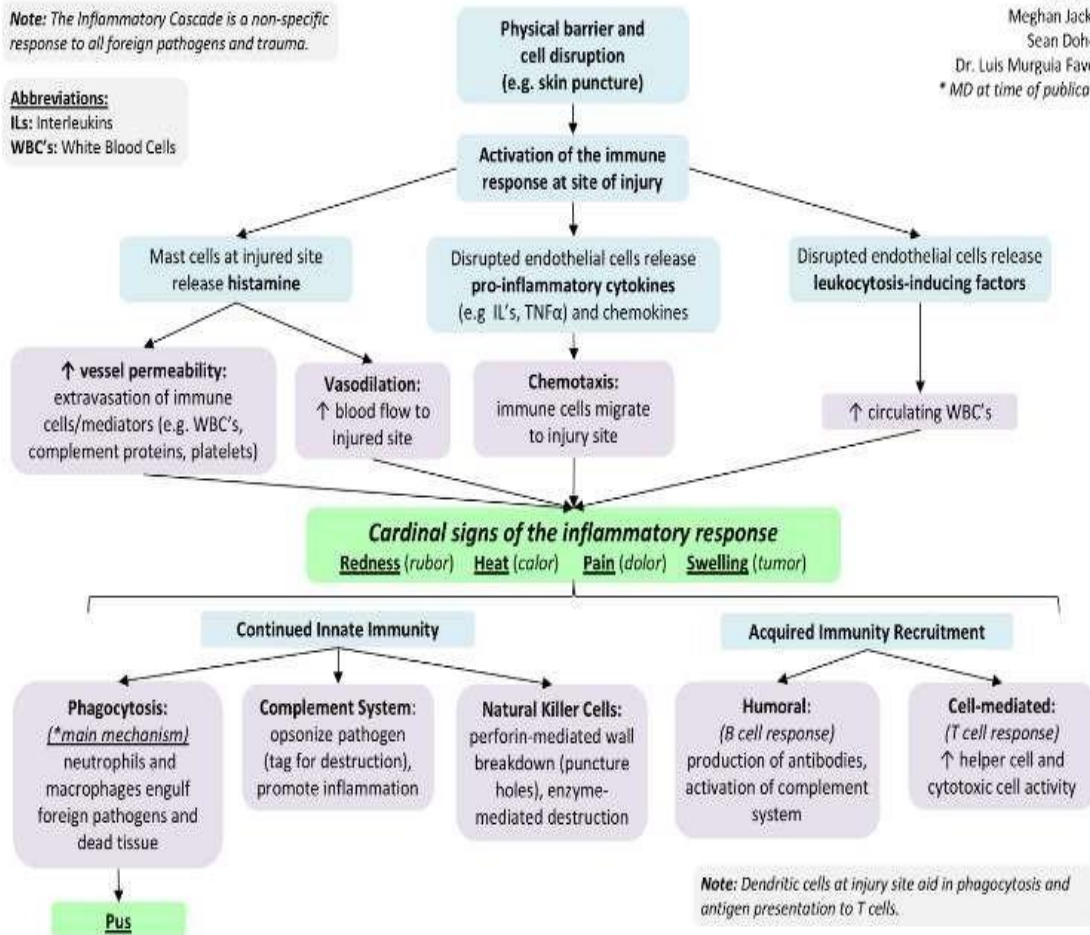


Figure 5a,b : Pathophysiology of Inflammation.

## Anti-inflammatory Drugs

### Synthetic Anti-inflammatory drugs

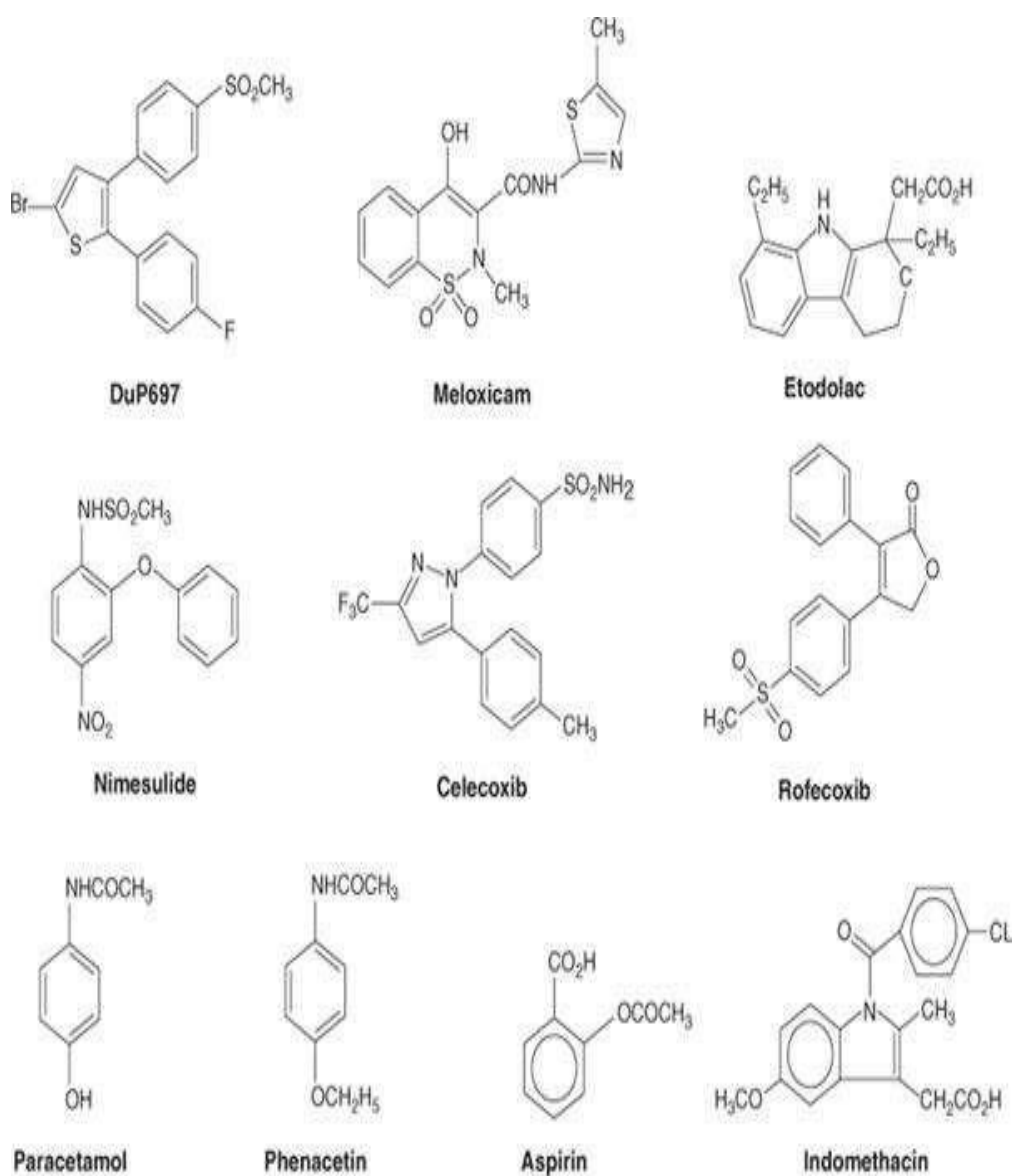


Figure 6: Synthetic Anti-inflammatory drugs (NSAIDs).

Virshette *et al.*, 2019, synthetic anti-inflammatory drugs are useful in treatment of acute and chronic inflammatory processes (Lima and Alvim, 2018). Pereira-Leite *et al.*, 2017, NSAIDs cover a wide spectrum of medications and their actions are all linked to COX inhibition in the generation of prostaglandins and thromboxanes (Sandoval *et al.*, 2017; Sostres and Lanas, 2016).

Wallace (2001), synthetic / allopathic anti-inflammatory drugs in high doses / prolonged therapy causes severe side effects like liver dysfunction, affect blood parameters and GIT disorders. (Shih and Chang, 2007; Calixto *et al.*, 2004)

### Medicinal Plants As Anti-inflammatory Agents

Laloo and Hemalatha (2011), India with its biggest repository (Medicinal Garden of the Globe; traditional herbal medicines / Alternative System of Medicine). Goyal *et al.*, 2003, natural products (NPs) can be considered any substance in the cosmos. Verma (2016), plants can synthesize a diverse range of phytochemical secondary metabolites.

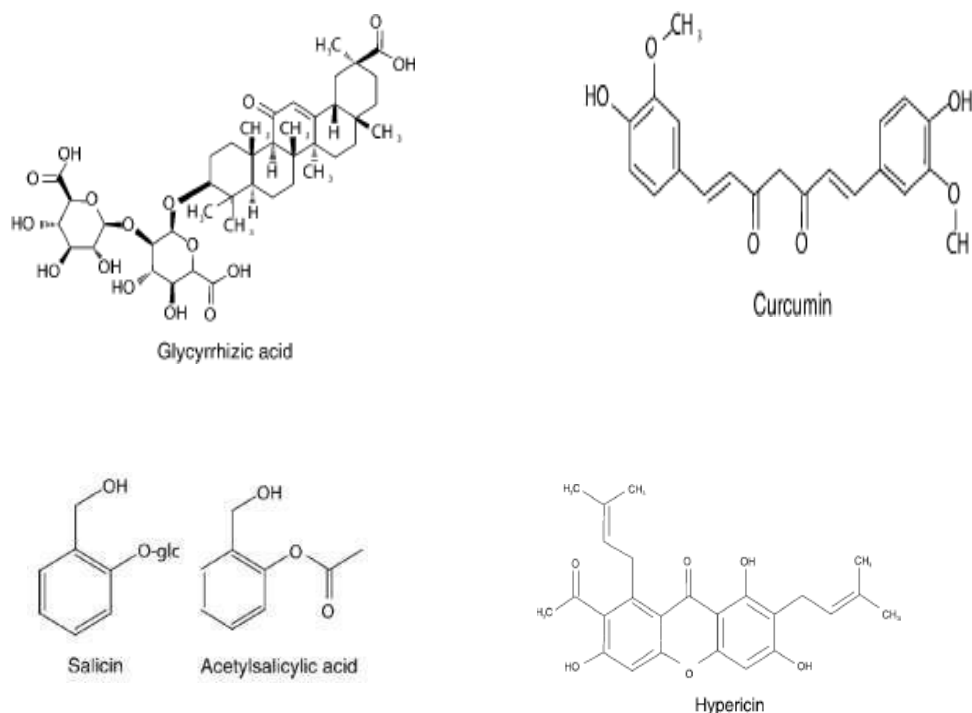


Figure 7: Natural Products as Anti-inflammatory Agents.

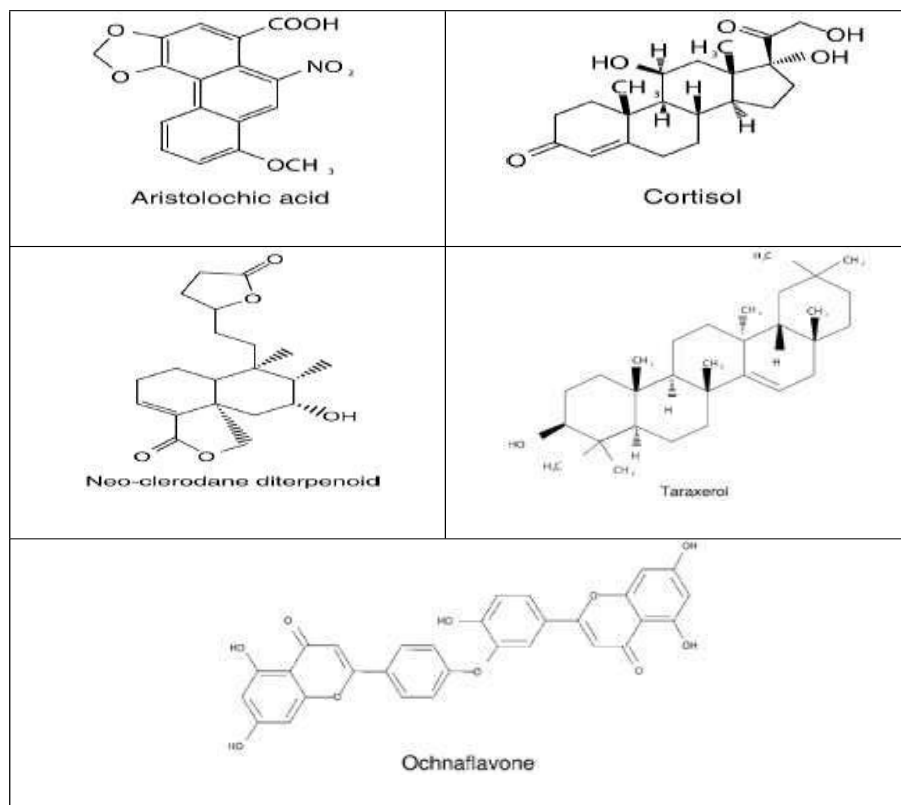


Figure 8: Natural Products as Inhibitors of Phospholipase A2.

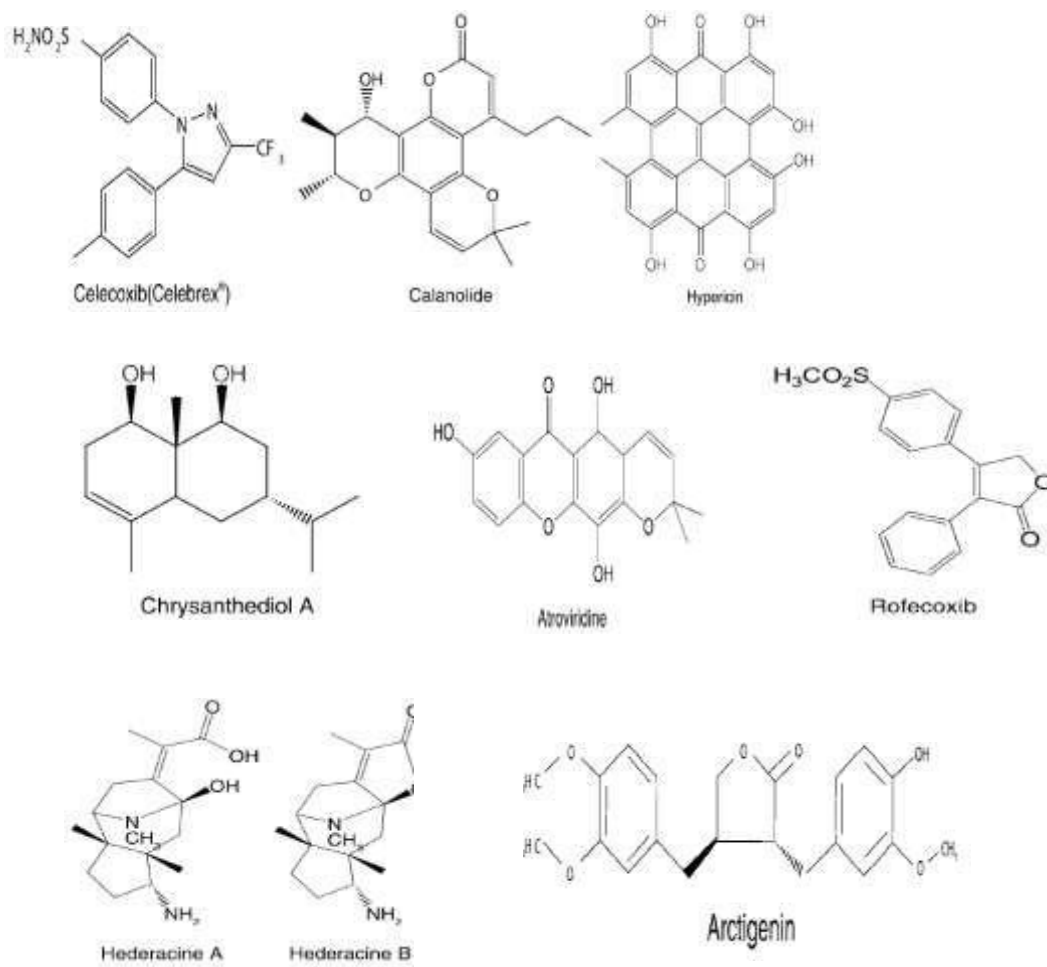


Figure 9: Natural Products as Inhibitors of COX.

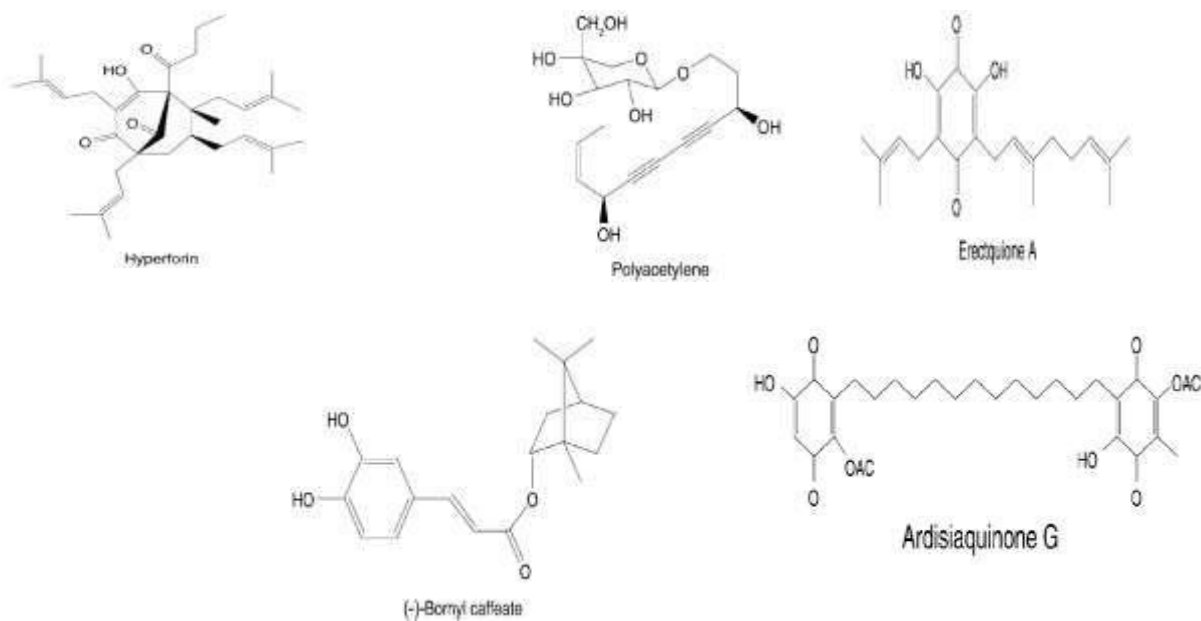


Figure 10: Natural Products as Inhibitors of Lipoxygenases.

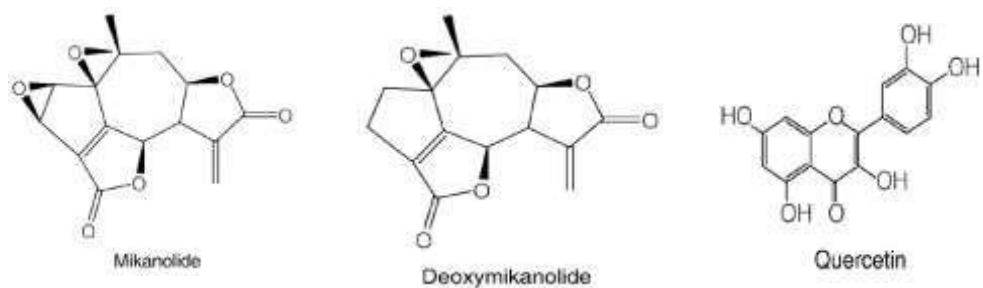


Figure 11: Natural Products as Inhibitors of Elastase.

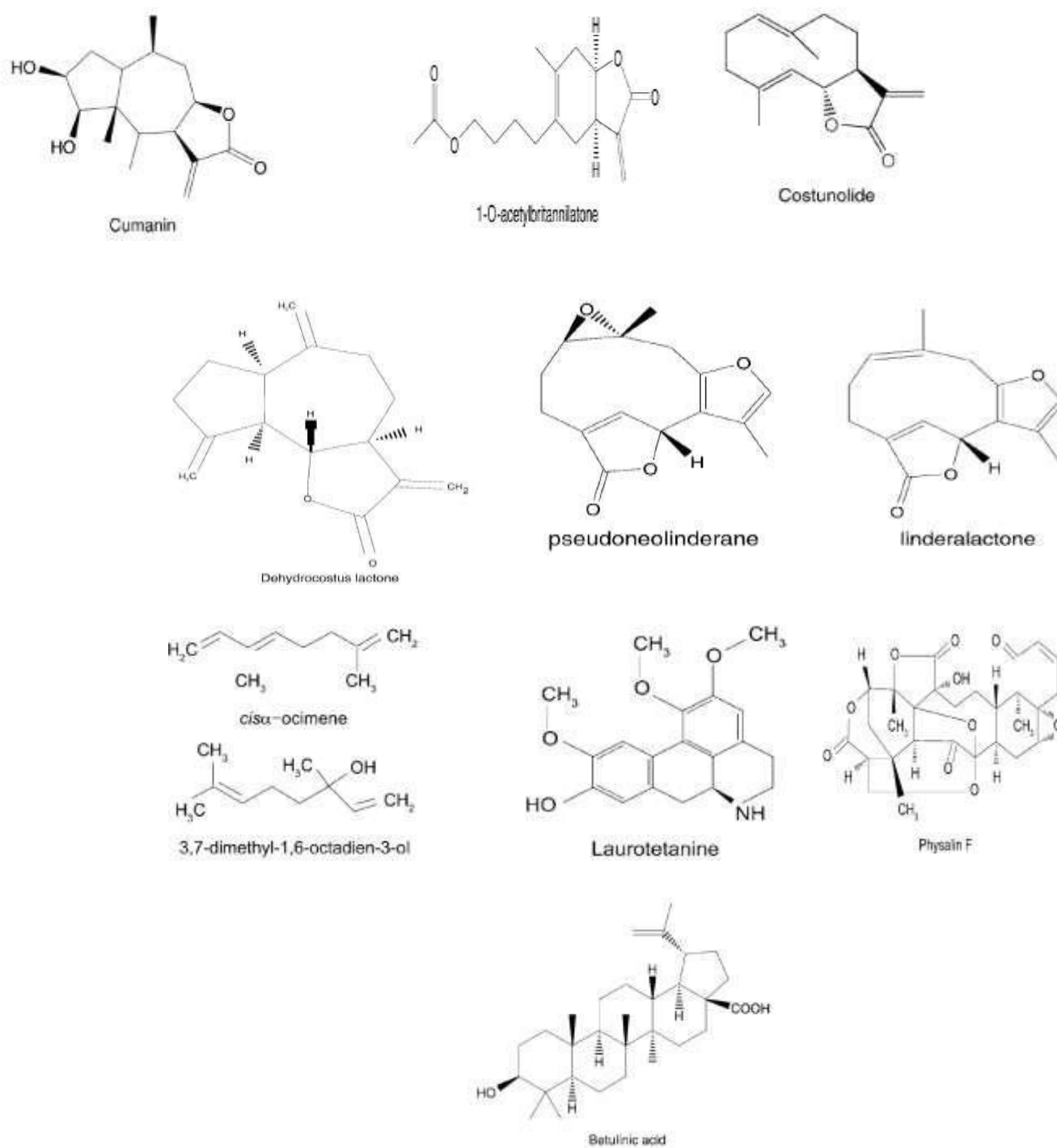


Figure 12: Natural Products as Inhibitors of Nitric Oxide Synthetase.



Table 1 : Medicinal plants with Anti-inflammatory activity.

Botanical / Family / Common Name	Parts Used	Constituents	Action / Uses
<i>Allium sativum</i> (Liliaceae) Lasun	Bulb, tuber, oil	Acrid volatile oil, starch, albumen.	Inflammation, anthelmintic, diuretic, carminative, antiseptic.
<i>Beta vulgaris</i> (Chenopodiaceae) Beet	Root leaves	Betin.	Carminative, emmenagogue, Diuretic, inflammation.
<i>Calotropis gigantea</i> (Asclepiadaceae) Rhui	Root, plant, flower, juice, bark	Glucosides, calotropin, mudarine.	Purgative, anthelmintic, Expectorant, inflammation, Stomachic, leukoderma.
<i>Cinnamomum zeylanicum</i> (Lauraceae)	Stembark	Resin, linalon, tarmin cinnamomum,	inflammation, stimulant, stomachic, diuretic.
<i>Coriandrum sativum</i> , Dhania (Umbelliferae)	Leaf, bark, mucilage of fruit	Tannin, catharin, albuminoids,	Carminative, anti- inflammation, jaundice, diuretic.
<i>Cuscuta reflexa</i> (Convolvulaceae)	Plant, seed, fruit, stem.	Cuscutine coumarin.	Inflammation, eye diseases, blood purifying.
<i>Ficus religiosa</i> (Moraceae) Pipala	Bark, leaves, fruits, seeds	tannins, rubber and wax.	inflammation, diarrhoea, stomatitis, hemorrhages.
<i>Foeniculum vulgare</i> (Apiaceae) Shepu	Fruit, root, seeds, leaves.	Estragole, coumaric-acid,	antitumor, antioxidant, anti-inflammatory,
<i>Gymnema sylvestre</i> (Asclepiadaceae)	Whole plant	gymnemic acid, tartaric acid,	Inflammation, bronchitis cardiotonic, laxative,
<i>Hibiscus rosasinensis</i> (Malvaceae) Jaswand	Buds, roots, leaves,	Quercetin, Ascorbic- acid.	anti-inflammatory, antibacterial, analgesic, astringent, cardiotonic.
<i>Justicia gendarussa</i> (Acanthaceae) Nilinirgundi	Roots, leaves.	Amino benzyl- Alcohol, Beta- sitosterol.	Anti-inflammatory, thermogenic, bronchitis, ascites, cough.
<i>Maytenus emargiata</i> , Yekaddi (Celastraceae)	Fruit, stem, bark, leaves, roots.	Tingenone, betulin, b- sitosteol.	Inflammation, ulcer, piles, burning, corneal opacity.
<i>Momordica charantia</i> , karela (Cucurbitaceae)	Whole plant	beta-carotene, niacin, momordicoside	anti-inflammatory, emetic, antidiabetic, appetizing, emmenagogue.
<i>Nelumbo nucifera</i> (Nymphaeaceae) Kamal	Whole plant.	hyperoside, d- catechin, rutin, trigonelline	cardiotonic, inflammation, leprosy, skin diseases, bronchitis, vomition
<i>Nicotiana tobacum</i> (Solanaceae) Tamabaku	Leaves.	nicotinic-acid, nicotine, tocopherol	anti-inflammatory, laxative, trigonelline, mental

<i>Nigella sativa</i> (Ranunculaceae)Kalajira	Seeds.	carvone, methionine, stigmaterol	Anti-inflammatory, carminative, thermogenic, emmenagogue, anodyne.
<i>Pterocarpus marsupium</i> (Fabaceae)	Heart wood, leaves	Alkaloids, gum, essential	Anti-inflammatory, anthelmintic, constipating
<i>Ricinus communis</i> (Euphorbiaceae)Arandi	Root, leaves, seeds,flowers, oil.	ricin, palmitin, sterine.	anthelmintic, diuretic, astringent, galactagogue, expectorant.
<i>Rubia cordifolia</i> (Rubiaceae)Manjeshtha	Roots.	Starch, colouring matter	Anti-inflammatory, carminative, diuretic, galactopurifier.
<i>Swertia chirayita</i> (Gentianaceae)Chirayita	whole plant	resin, gum, resin, phosphate	Anti-inflammatory, antipyretic, thermogenic, antiperiodic.
<i>Tamarindus indica</i> (caesalpiniaceae)Chinch	Roots, leaves,Fruits	Tartaric, citric, malic, acetic,	Astringent, thermogenic, constipating, diuretic, stomachic
<i>Taraxacum officinale</i> (Asteraceae)	Whole plant	Latex contain taraxacerin. pectin	Anti-inflammatory, thermogenic, digestive, stomachic, stimulant.
<i>Terminalia arjuna</i> (Combretaceae)	bark	Not reported	Anti-inflammatory, astringent, dysenteric.
<i>Terminalia belirica</i> (Combretaceae)	Bark, fruits / Beheda	Not reported.	Anti-inflammatory, thermogenic, astringent.
<i>Terminalia chebula</i> (Combretaceae)Hirda	Mature, immature fruits.	Gallic acid, Ellagic acid, Chebulic acid.	anti-inflammatory, purgative, antiseptic, diuretic, cardiotonic.
<i>Tinospora cordifolia</i> , Giloi (Menispermaceae)	Stem	Alkaloids,starch.	anti-inflammatory, antiemetic, expectorant, digestive.
<i>Tribulus terrestris</i> (Zygophyllaceae)Gokhru	Whole plant	Diuretics.	Anti-inflammatory, laxative, appetiser, styptic, diuretic
<i>Vitex negundo</i> (Verbenaceae)Nirgundi	Whole plant	essential oil, resin, astringent.	Expectorant, anti- inflammatory, digestive, antipyretic,
<i>Zingiber officinale</i> (Zingiberaceae)Adrak	Rhizomes	camphene, cineol, shogaol, zingiberene,	anodyne, expectorant, anthelmintic, carminative, thermogenic.

Virshette *et al.*, 2019, non-steroidal anti-inflammatory drugs (NSAIDs) are used to treat inflammation (acute and chronic pain) by inhibition of COX-1 and COX-2 which stop accumulation of prostaglandins and thromboxanes (Lima and Alvim, 2018; Pereira-Leite *et al.*, 2017; Sandoval *et al.*, 2017).

Wallace (2001), NSAIDs in prolonged therapy causes deleterious side effects such as gastric lesions, cardiovascular, renal and gastrointestinal damage. Percival (1999), disadvantage of NSAIDs is their toxicity and reappearance of symptoms after discontinuation. So, screening and development new anti-inflammatory drugs are the need of hour and efforts are made to find natural products (NP) as anti-inflammatory drugs.

Shih and Chang (2007), more than 80% of medicines have been developed from natural products (NP) obtained from natural source. Medicinal plants play an important role in the development of potent therapeutic APIs. Indian System of Medicine and India is considered as biggest repository of medicinal plants or “Medicinal Garden of the Globe”. Stimulation of the immune system, regulation of gene expression in cell proliferation and apoptosis, antibacterial and antiviral effects.

Many natural products (NPs) and herbal formulations having anti-inflammatory activity are patented. Anti-inflammatory activities of Ethanolic Extract of roots of *Asparagus racemosus* Willd (EERAR) against carageenan was undertaken with following objectives :

- Qualitative analysis of *Asparagus racemosus* Willd. (Shatavari) roots
- Toxicity studies of EERAR;
- *In-vitro* and *in-vivo* anti-inflammatory studies of EERAR;
- Pharmacodynamics of EERAR;

#### Methodologies Used:

- phytochemical screening and chromatography;
- Biochemical analysis;
- Anti-inflammatory activity against Carageenan.

#### *Asparagus racemosus* Willd

Goyal *et al.*, 2003, genus *Asparagus* consists of more than 250 species (22 species in India) distributed in tropical and subtropical regions throughout the globe (at low altitudes in shade throughout Asia, Australia and Africa). *Asparagus racemosus* (Shatavari or Sanspayein) is used since Pre-Vedic times (mentioned ayurvedic literature 5000 years ago) belongs to genus *Asparagus* (Asparagaceae; Liliaceae) and habitat of tropical and subtropical regions (Simon, 1997).

Shashi *et al.*, 2013, shatavari means “curer of a hundred diseases and it is a general tonic as well as a female reproductive tonic (rejuvenative tonic for female). It is known as Queen of herbs with ability to increase fertility and vitality (translated as 100 spouses).

#### Pharmacognosy of *Asparagus racemosus* Willd.

Table 2: Pharmacognostical details of *Asparagus racemosus* Willd.

Foliage	:	Evergreen
Leaf type	:	Phylloclades (photosynthetic branches), uniform;
Leaf colour	:	Shiny green
Leaf surface	:	pine-needle, shiny and glossy surface;
Type of stem	:	Woody;
Roots	:	Adventitious root system with tuberous roots;
Flower	:	minute, white flowers;
Fruit	:	Greenish (unripe) to red (ripe) to blackish-purple (dried), globular berries;



Figure 13 : Field Photograph of *Asparagus racemosus* Willd.



Figure 14 : Photograph of flowers of *Asparagus racemosus* Willd.



Figure 15 : Photograph of Roots of *Asparagus racemosus* Willd.

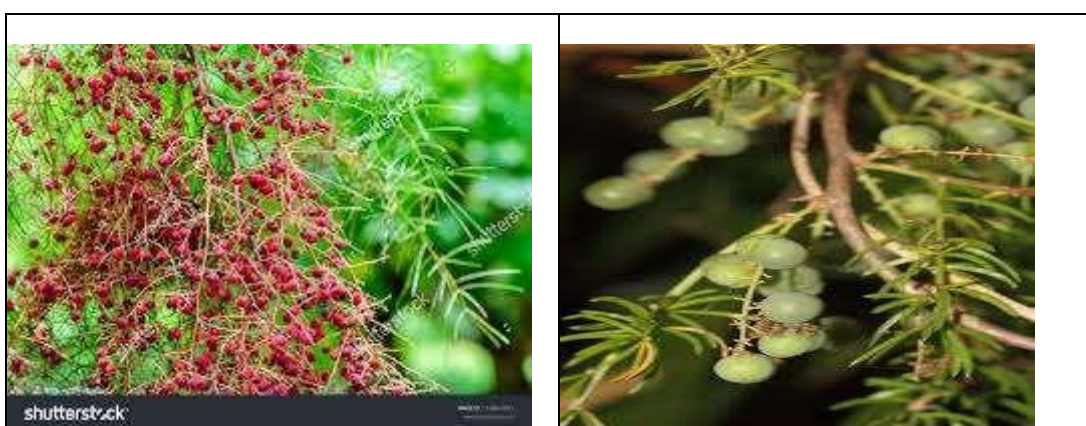


Figure 16: Photograph of ripe and unripe fruits of *Asparagus racemosus* Willd.



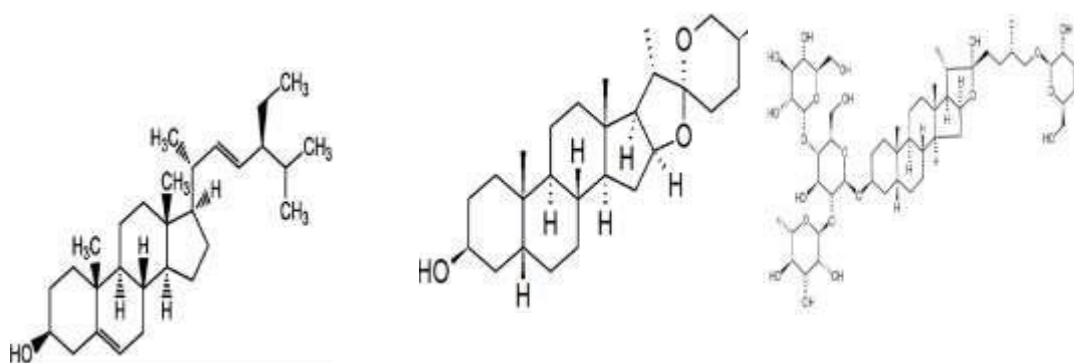
Figure 17: Photograph of Complete plant of *Asparagus racemosus* Willd.

### Characteristics of *Asparagus racemosus* Willd

Table 3: Description of *Asparagus racemosus* Willd plant parts.

Plant	:	under-shrub, spinous, short root stocks
Leaves	:	resemblance with pine needles
Root	:	Elongated, tuberous (brown), tapering ends, 1-2 cm in thick;
Flowers	:	Uniform, white, small spikes, hermaphrodite, aromatic,
Fruit	:	red berries, small, round / globular in shape, 2 to 3 lobed,

### Chemistry of *Asparagus racemosus* Willd.



Stigmasterol

Sarsasapogenin

Shatavarin IV

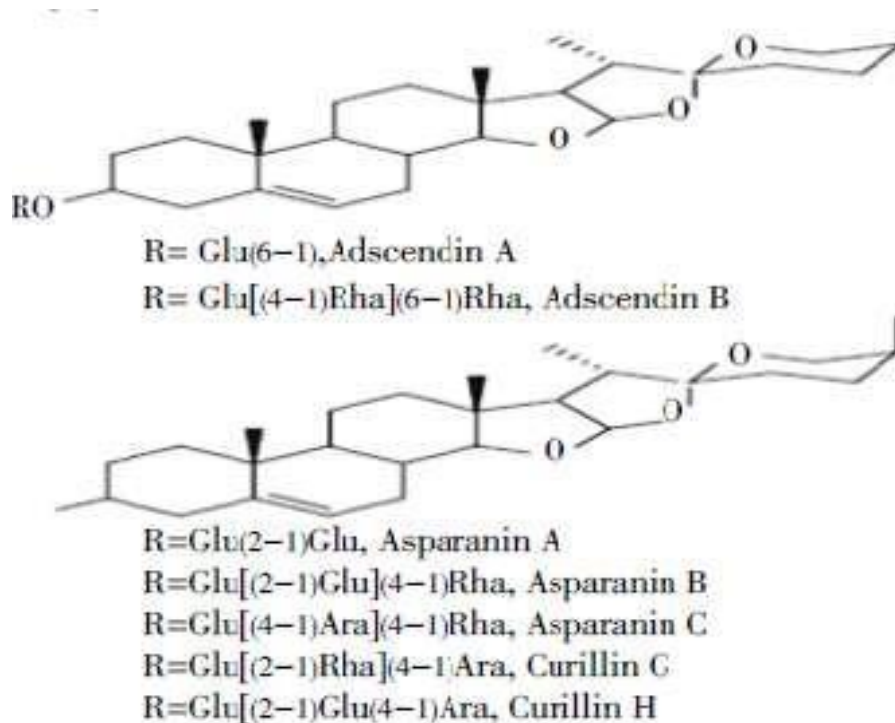


Figure 18: Structure of compounds present in *Asparagus racemosus* Willd.

**Traditional and Pharmacological uses of *Asparagus racemosus* Willd.**Table 4: Ethnopharmacological & Pharmacological uses of *Asparagus racemosus*.

Ethno- pharmacological: Uses	➤ maintains female hormonal balance ➤ nourish female reproductive organs ➤ ageing (increase longevity) ➤ immunity builder, improve mental function, ➤ vigor and addvitality to the body ➤ nervous disorders, dyspepsia, ➤ inflammation, neuropathy, hepatopathy
Pharmacologicaluses	: Anti-abortifacient; anti-ulcerogenic; anti-oxidant; anti-diabetic; anti-periodic; anti-neoplastic; immune-modulatory; adaptogenic; anti-inflammatory; anti-bacterial, anti-leprotic. (Lakhwinder <i>et al.</i> , 2018)

Table 5 : Pharmacological activities of *Asparagus racemosus* Willd.

Activity	Reference
Adaptogenic	Forinash <i>et al.</i> , 2012;
Antioxidant	Palanisamy <i>et al.</i> , 2012
Cardio-protective	Deepika and Dimple (2014)
Anti-ulcer	Bhatnagar <i>et al.</i> , 2006;
Antimicrobial Anti-bacterial	/ Jagannath <i>et al.</i> , 2012 (against <i>Vibrio cholerae</i> , <i>Staphylococcus aureus</i> , <i>Escherichia coli</i> , <i>Salmonella typhi</i> )
Reproductive	Somania <i>et al.</i> , 2012

**Materials and Methods****Procurement and Authentication of *Asparagus racemosus* Willd**

Fresh roots of *Asparagus racemosus* Willd. (Asparagaceae) were harvested locally from the herbal garden of the IEC Institute in the month of October 2022. Harvested *Asparagus racemosus* Willd. (Shatavari) roots were evaluated macroscopically, microscopically and chemical tests.

**Morphology and Microscopy of Roots of *Asparagus racemosus* Willd**

- Elongated, tuberous (brown); 1-2 cm in thick, 25-90 cm long;
- tapering ends, adventitious and tuberous roots;
- silver ash white internally / externally;
- pitted parenchyma cells with or without the inter-cellular spaces;
- starch grains present; Aseptate fibre, septate fiber, tracheid;
- piliferous layer with root hairs; unicellular root hair;
- cortex, raphide bundle and sap cell content;
- acicular rapids, rapid bundles and pericyclic fibres;

### Phytochemical screening of roots of *Asparagus racemosus Willd.*

Roots of Shatavari (*Asparagus racemosus Willd.*) were analysed for various phyto-constituents / nature of chemical groups using standard procedures described by Harborne (1973); Trease and Evans (1985); Sofowora (1993); Khandelwal (2008); Kokate (2005) (Table 6)

Table 6: List of qualitative chemical tests.

PPMs / SPMs	Chemical Test
Alkaloid	a. Mayer;
	b. Dragendorff;
Carbohydrate	Molisch's Reagent Test;
Reducing Sugar	a. Fehling's Reagent Test;
	b. Benedict's Reagent Test;
Saponins	a. Foam Test;
	b. Forth Test;
Cardiac Glycosides	Killer-Killani Test;
Proteins & Amino Acids	a. Millon's Reagent Test;
	b. Biuret's Test;
	c. Ninhydrin Reagent;
Terpenoids	Salkowski's Test;
Fixed Oils and Fats	a. Spot Test;
	b. Saponification Test;
Gum and Mucilage	Ruthenium Red Solution Test
Glycosides	Liebermann's Test;

### Preparation of EERAR extract of *Asparagus racemosus Willd*

- Freshly harvested roots of *Asparagus racemosus* was dried under shade;
- Coarsely pulverized shatavari roots (1000 kg) was subjected to Soxhlet extraction (continuous hot extraction method) with crude ethanol (80%) for 18 hours;
- Extracts filtered, distilled, concentrated, (vacuum evaporated ; 50°C);
- Concentrated extract was finally lyophilized;
- Practical yield EERAR extracts was 78.6 gm (7.86%);
- Different concentrations of EERAR extract (200 / 400 mg in 1% carboxymethyl-cellulose) were used in pharmacological studies;

### Physico-chemical Analysis of *Asparagus racemosus Willd*

#### Physico-chemical Parameters Analysis

In the experiment freshly procured, washed, air-dried and pulverised / powdered roots of shatavari was subjected to physico-chemical analysis for parameter like foreign matter, extractive values, swelling index, ash value (acid insoluble/water soluble), fluorescence analysis in various solvents (normal light/UV light), and loss on drying (Table 7 & 8) etc.

Table 7: Physicochemical analysis of roots of Shatavari.

Quantitative parameter	Values obtained (%) w/w
Alcohol Soluble Extractive Value (ASEV)	7.94±0.72
Water Soluble Extractive Value (WSEV)	8.62±0.46
Total Ash Value (TAV)	6.32± 0.24
Acid Insoluble Ash Value (AIAV)	1.6±0.08
Water Soluble Ash Value (WSAV)	4.42±0.18



Sulphated Ash Value (SAV)	2.12±0.14
Swelling Index (SI)	NIL
Loss on Drying (LOD)	2.68±0.04
Foreign Matter Content (FMC)	0.78%

Table 8: Physicochemical parameters of roots of shatavari.

Solvent used	Observation	
	UV light (200 nm)	UV light (400 nm)
Benzene	Light Green	Light Brown
Acetone	Yellow	Light Brown
Chloroform	Yellowish Green	Yellowish Brown
Etanol	Dark Yellow	Brown
H <sub>2</sub> O (Distilled)	Yellow	Light Brown
Dil. HNO <sub>3</sub> Soln.	Green	Bluish Green
Dil. H <sub>2</sub> SO <sub>4</sub> Soln.	Green	Dark Green
Conc. HCL Soln.	Yellowish Green	Yellowish Brown
Aq. NaOH solution	Green	Brown
NaOH in CH <sub>3</sub> OH	Light Green	Yellowish Brown

#### Anti-inflammatory activity of EERAR of *Asparagus racemosus* Willd.

##### Rat red blood cell stabilization method (*in-vitro* analysis)

As result of SOP, EERAR of *Asparagus racemosus* Willd induced powerful RRBC membrane stabilizing effect (45.54% to 86.26%) (Table 9).

Table 9: Membrane stabilization by EERAR.

EERAR Concentration (mg/ml)	Percentage of inhibition%	
20	33.10 +0.98	45.24 + 0.42
40	37.31 +0.34	51.32 +0.26
60	51.39 +0.18	65.10 +0.24
80	59.15 +0.92	79.03+ 0.12
100	66.47 +0.44	86.56+ 0.22

#### Carrageenan Induced Rat Paw Edema

Protocol was approved by IAEC Form B IEC/IAEC/ 2023/01 dated 17-02-2023.

Table 10 : Grouping for anti-inflammatory activity (6/group).

Group	Treatment / Dose
I	Normal Control: Water, ad libitum

II	Toxic Control : 0.1 mL of 1% carrageenan in 0.9 % NaCl subcutaneously (s.c.);
III	Carrageenan + EERAR-A (200 mg/kg p.o.);
IV	Carrageenan + EERAR-B (400 mg/kg p.o.);
V	Carrageenan + Standard (10 mg/kg p.o)

SOPs were followed and paws were measured using Zeitlin's apparatus. All doses (suspension of EERAR extract of *Asparagus racemosus* Willd. in 0.5% sodium CMC; EERAR-A: 200 mg/kg/b.wt. or EERAR-B: 400 mg/kg/b.wt.) were administered orally 1 hr prior to induction of oedema. 0.1 mL carrageenan was then injected. Paw edema was measured at 1, 2, 3, 4, 5 and 6 hr percentage of paw edema inhibition was calculated.

Indomethacin (10 mg/kg/bw) was used as Standard and given orally followed by injection of 1% Carrageenan. Subsequently, percentage of paw edema inhibition was calculated by the formula:

$$\% \text{ Inhibition in paw thickness} = \frac{(Y_t - Y_0) \text{ control} - (Y_t - Y_0) \text{ treated}}{(Y_t - Y_0) \text{ control}} \times 100$$

Where,

$Y_0$  = hind Paw volume at 0 hr.

$Y_t$  = hind Paw volume at time t (t = 1, 2, ....6) after injection

Table 11: Effect of EERAR on paw edema.

Group	Mean Edema Thickness (MET) (time in hrs)					
	1	2	3	4	5	6
I (Normal)	--	--	--	--	--	--
II (Carrageenan)	0.23 ±0.03	0.35 ±0.05	0.46 ±0.05	0.58 ±0.05	0.65 ±0.06	0.62 ±0.10
III (EERAR-A: 200 mg/kg)	0.23 ± 0.03	0.30 ± 0.02	0.38 ± 0.02	0.47 ± 0.04	0.53 ± 0.06	0.33 ± 0.06
IV (EERAR-B: 400 mg/kg)	0.18 ±0.03	0.30 ±0.00	0.35 ±0.03	0.41 ±0.03	0.45 ±0.03	0.26 ±0.04
V (Indomethacin; 10 mg/kg)	0.15 ±0.03	0.22 ±0.04	0.28 ±0.04	0.33 ±0.04	0.38 ±0.02	0.21 ±0.05

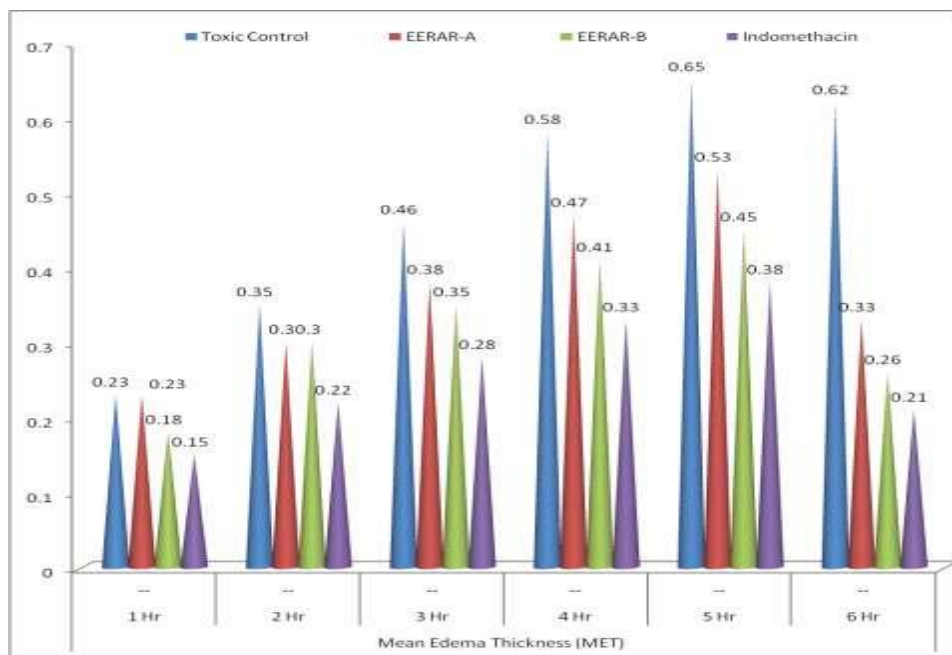


Figure 19: Effect of EERAR on edema.

Table 12: Inhibition of paw edema.

Group	% Inhibition (after 1-6 hrs)					
	1	2	3	4	5	6
I :Normal	--	--	--	--	--	--
II : Carrageenan	--	--	--	--	--	--
III : EERAR-A	7.86	10.98	13.48	17.58	20.92	22.24
IV : EERAR-B	10.34	14.92	19.68	24.56	28.22	32.78
V : Indomethacin	13.22	16.82	21.28	27.34	31.92	35.72

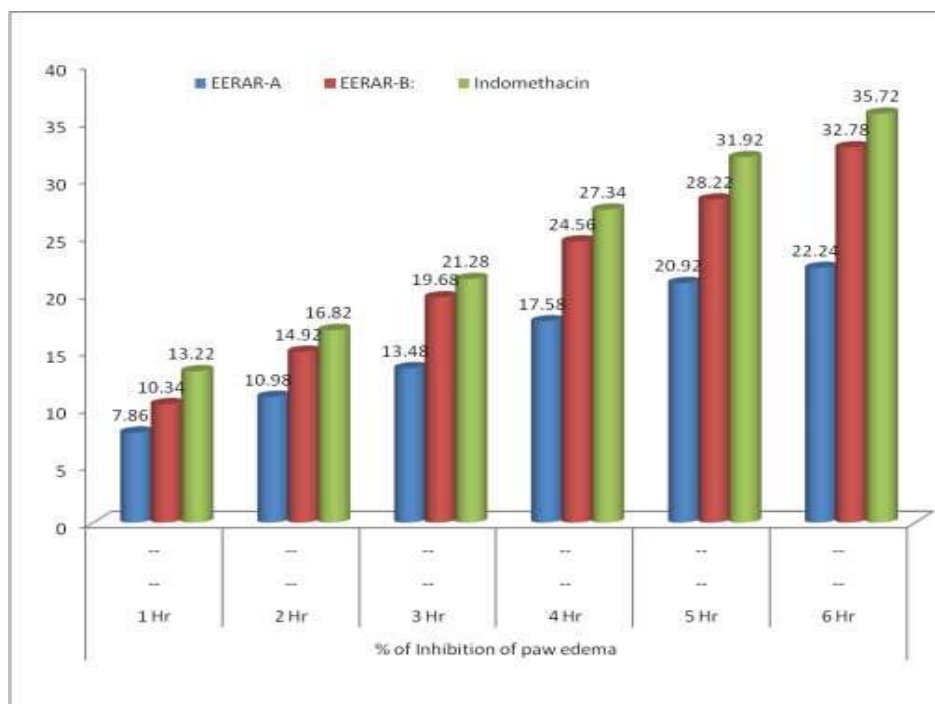


Figure 20: EERAR induced % inhibition of paw edema.

**Results and Discussions**

Pharmacognostical analysis of *Asparagus racemosus* Willd. (shatavari) roots confirmed the authenticity and herbarium specimen was deposited in Herbarium Bank (IEC/Pharm/Herb/2022/2224). Besides, chemical tests analysis of EERAR detected

phenolic compounds, flavanoids, steroidal glycosides reducing sugars, amino acids, isoflavones, tannins, carbohydrates, coumestans, and rutin and traces of minerals (Zinc, manganese, copper, calcium, magnesium, potassium).

In physicochemical analysis of roots of *Asparagus racemosus*, ash values were found to be very low (roots were free from contamination). Low acid-insoluble ash values AIAV 1.6 % indicated that the drug was free from siliceous matters or sand. Water soluble ash value WSAV 4.42 % indicated presence of water soluble salts. Solvent extractive values of ASEV (7.94 %) and WSEV (8.62%) showed presence of phyto-constituents and LOD was 2.68% (due to moisture content).

The EERAR extract of *Asparagus racemosus* Willd demonstrated powerful RRBC membrane stabilizing property (45.24% to 86.56%). To induce edema 1% carrageenan was administered into right hind paw and induced paw edema was measured using Zeitlin's apparatus (1,2,3,4,5 and 6 hr after induction of edema). In assessment of anti-inflammatory activity, group-III & group IV (drug / EERAR extract Treated; (suspension of EERAR extract of *Asparagus racemosus* Willd. in 0.5% sodium CMC) were administered orally 1 hr prior to induction of edema followed by carrageenan and paw thickness measurements were recorded after each hour for 6 hrs and %age inhibition of paw edema. Group V was administered Indomethacin (10 mg/kg/bw.p.o.) followed by injection of 1% carrageenan and inhibition of paw edema. It was found that EERAR (A/B) induced anti-inflammatory activity against the phlogistic agent. EERAR-B produced better anti-inflammatory effect than EERAR-A. The standard drug indomethacin (a known cyclooxygenase inhibitor) induced excellent anti-inflammatory and anti-edematous effects (edema reduced significantly). Anti-inflammatory effects of indomethacin and EERAR-B (400 mg/kg/b.wt.) were very good in comparison of drug vehicle treated group (values were significant at  $P < 0.05$  over control). EERAR contains flavonoids which produced significant inflammatory effects. Comparatively, anti-edematous and anti-inflammatory effects of EERAR-B extract were lesser than the Standard drug – indomethacin.

### Conclusions

*Asparagus racemosus* roots were authenticated and phenolic compounds, alkaloids, sterols, flavanoids, steroidal glycosides isoflavones and tannins were found present. In physicochemical analysis of roots showed AIAV (1.6 %), WSAV (4.42 %), ASEV (7.94 %), WSEV (8.62%) and LOD (2.68%). *Asparagus racemosus* EERAR showed wide safety margin, classified as Non-Toxic Constituent, LD50 value was 2000 mg/kg b.wt. did not caused any significant change in blood, liver and kidney parameters with slight abnormal behavior. EERAR extract induced powerful Rat RBC membrane stabilizing property (45.24% to 86.56% membrane stabilization impact of EERAR). Anti-inflammatory activity was maximum in standard (35.72%) followed by EERAR-B (400 mg; 32.78%). EERAR-B produced better effect than EERAR-A (significant at  $P < 0.05$  over control).

### REFERENCES:

- Bhatnagar, M., Sisodia, S. (2006). *J. Herb. Pharmacother.* 6(1), pp. 13-20.
- Burke A., Smyth E., & FitzGerald, G.A. (2005). *Analgesicantipyreticagents*. In: Goodman & Gilman. pp. 671-715.
- Calixto, J.B., Otuki, M.F., Santos, A.R.S.(2004). *Planta Medica.* 70, pp.93-103.
- Campos, J.F., Macorini, L.F.B., Balestieri, J.B.P, (2014). *Food andChemical Toxicology.* 65, pp. 374-780.
- Deepika, C., Dimple, S.A. (2014). *Int. J. Sci. Res.* 3(7), pp.742-46.
- Forinash, A.B., Yancey, A.M., Barnes, K.N. (2012). *Ann Pharmacother.*46(10), pp. 1392-404.
- Goyal, R.K., Singh, J., Lal, H. (2003). *Indian J Med Sci.* 57(9), pp. 408-414.
- Harborne, J. B. (1973). *Phytochemical Methods: A Guide to Modern Tech.of Plant Analysis*. Springer Publications. pp. 1-288.
- Jagannath, N., Somashekara, S.C., Damodaram, G., Devasankaraiah, G.(2012). *Indian J Pharmacol.* 44(5), pp. 576-79.
- Khandewal, K.R. (2008). *Prac. Pharmacognosy*. Nirali, ed.19, pp. 1-67.
- Kokate, C. K. (2005). *Practical pharmacognosy*. 5th Edition, VallabhPrakashan: New Delhi. pp. 107- 111.
- Lakhwinder, S., Antul, K., Anuj, C. and Gurwinder, S. (2018). *Journal ofPharmacognosy and Phytochemistry* 7(3), pp. 2199-2203
- Laloo, D., Hemalatha, S. (2011). *Pharmacogn Rev.* 5, pp. 147-54.
- Libby, P. (2003). Harvard Medical School, Brigham Boston. Pp. 56-67.
- Lima, A.S., Alvim, H.G. (2018). *Revista de Iniciacao Cientifica eExtensao.* 1, pp. 169-174.
- Palanisamy, N., Manian, S. (2012). *Toxicol Ind Health.* 289(3), pp. 238-44.
- Parhnam, M., and Battista, J.A. (2008). *J. of Int. Assoc.of Inflamm. Soc.* pp. 1023-1030.
- Percival, M. (1999). *Clinical nutrition insights.* 4, pp. 1-5.
- Pereira, C, Nunes, C., Jamal, S.K., Cuccovia, I.M. (2017). *MedicinalResearch Reviews.* 37 (4), pp. 802-859.
- Sandoval, A.C., Fernandes, D.R., Junior, T.A. (2017). *Revista Cientifica daFaculdade de Educacao e Meio Ambiente.* 8(2), pp. 165-176.
- Shashi, A., Sanjay, K., Amita, V., Mayank, K., Monika, S. (2013). *AsianPacific Journal of Tropical Disease.* 3(3), pp. 242-251.
- Shih, S.C., Chang, C.W. (2007). *Int. J. of Gerontology.* 1 (1), pp. 40-45.
- Simon, D. (1997). *The wisdom of healing*. Harmony Books. pp. 148-157.
- Sofowora, A. J. (1993). *Altern. Complement. Med.* 2(3), pp. 365-372.
- Somania, R., Singhai, A.K., Shivgunde, P., Jain, D.P. (2012). *Indian J ExpBiol.* 50(7), pp. 469-475.

26. Sostres, C., Lanas, A. (2016). *Medicina Clínica*. 146(6), pp. 267-272.
27. Trease, G.E., Evans, W.C. (1985). *Pharmacognosy*. 17th edn., Bahiv Tinal, London: pp, 149.
28. Verma, S. (2016). *Journal of Phytopharmacology*. 5(4), pp. 157-159.
29. Virshette, S.J., Patil, M.K., Somkuwar, A.P. (2019). *Phytochemistry*. 8(4), pp. 1641-1646.
30. Wallace, J.L.(2001). *Best Practice and Res. Clin.Gastroenterology*. 15(5), pp. 691-703.