Chapter 8

Anti-inflammatory Evaluation of *Meyna laxiflora*
8.1. Introduction

8.1. 1. Inflammation and its types

Inflammation is a pervasive form of defense that is broadly defined as a nonspecific response to tissue malfunction and is employed by both innate and adaptive immune systems to combat pathogenic intruders.\(^1\) Inflammation, in its broadest sense, is a host response to tissue injury which involve defense reaction resulting elimination of injurious agent necrosed cells or tissue. The four ancient, cardinal, signs of inflammation described by Roman writer Celsus are redness, heat, swelling, and pain while loss of function was described by Virchow.\(^2,3\)

8.1. 2. Causative agents\(^3\)

- Infective agents (bacteria, viruses, fungi, parasites etc.).
- Immunological agents (cell mediated and antigen-antibody reactions).
- Physical agents (heat, cold, radiation, mechanical trauma etc.).
- Chemical reagents (Organic and inorganic poison).
- Inert material (foreign particle like dust, pollen etc.).
8.1. 3. Classification of inflammation

Depending upon the defense capacity of host and duration response inflammation may classify as acute and chronic inflammation.³

❖ Acute Inflammation

Acute inflammation is the immediate and early response to injury lasting less than two week which involve leukocytes and plasma proteins as a mediator of host defense. It is characterized by following (Figure 8.1)

➢ Alteration in vascular caliber that lead to increase in blood flow.
➢ Elimination of plasma protein and leukocytes from circulation and accumulation at site of inflammation.
➢ Activation of polymorphonuclear neutrophils for elimination of offending agent.

❖ Chronic Inflammation

Chronic inflammation can be considered to be inflammation of prolonged duration (weeks to months to years in which active inflammation, tissue destruction, and attempt for healing of injurious cell proceed simultaneously. It occurs if causative agents of acute inflammation persist for long time, autoimmunity and exposure of potentially toxic substance either exogenous or endogenous. It is characterized by following (Figure 8.2)

➢ Infiltration with mononuclear cells which including macrophages, lymphocytes, and plasma cells
➢ Tissue destruction induced by offending agent or, inflammatory cells
➢ Attempt of healing at connective tissue replacement of damage tissue accomplished by angiogenesis and fibrosis.
Figure 8.1. Mechanism of acute inflammation

Figure 8.2. Mechanism of chronic inflammation
8.1.4. Pathogenesis

Inflammation is combination of various immunological, physiological, and behavioral processes that are organized by soluble immune signaling molecules called cytokines. The pathogenesis of inflammation depend upon following steps.¹ (Figure 8.3)

- Recognition of infection or damage
- Activation of common signaling pathways
- Release of pro-inflammatory cytokines
- Recruitment of effector cells
- Polarization of Inflammation
- Resolution of Inflammation

Initiators of inflammation such as pathogens, environmental factor, physical factors etc when comes in contact with cells or tissue they produce injury to them which recognized with the help of innate immune system and pathogen associated molecular patterns (PAMPs), which are specifically directed toward general structures of molecules expressed by pathogens that are essential for pathogen survival.

Many damage signals are recognized by germ-line encoded receptors, such as trans membrane Toll-like receptors (TLRs) and intracellular nucleotide binding domain (NOD-like receptors) and nucleotide binding leucine-rich-repeat- containing receptors (NLRs) which activate common signaling pathways responsible for release of NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells) from inhibitor protein, IκB in all type of cells. Activated NF-κ B binds with target genes in nucleus and produce transcription.

Transcription and translation of genes lead to release of pro-inflammatory cytokines, such as interleukin-1-beta (IL-1β), IL-6, tumor necrosis factor-alpha (TNF-
α), and others. The release of these chemokines facilitates the recruitment of effector cells, such as monocytes and neutrophils, to the site of disturbance in conjugation with various co stimulatory molecules. These cells produce highly reactive oxygen and nitrogen species and various proteinases which destructive to both pathogens and hosts and essentially induce liquefaction of surrounding tissue to protect from microbial metastasis and initiate conventional cardinal signs of local inflammation: heat, swelling, redness, pain, and loss of function.

These cells then migrates to prime naive T cells (Th0; never exposed to antigen) in lymphoid tissue and bound to MHC class II receptors. After that naive T cells produce different types of effector and regulatory cells: Th1 and Th17 cells (pro-inflammatory), Th2 cells (anti-inflammatory) and regulatory T-cells (Tregs). Th1 cells and Th17 cells produces cytokines (IFN-γ, IL-2, NF-α) (IL-17, IL-6, NF-α) respectively which are important for initiation of delayed type hypersensitivity responses and macrophage activation. Th2 cells produce a different characteristic set of anti-inflammatory cytokines, such as IL-4, IL-5, IL-10, and IL-13 that promote alternative activation of macrophages, and responsible for conversion of B-cell antibody to IgE and eosinophil maturation, while down regulating it produce Th1 cytokines hence responsible for inflammation. Treg cells secrets IL-10 and TGF-β which regulate homeostasis of the immune system by moderating Th1 and Th2 responses.

The last phase of inflammation is its resolution which starts after few hours of inflammation lipoxin produced by macrophages which block further neutrophil recruitment and increases uptake of monocytes which responsible for wound healing.
8.1. 5. Diagnosis

Redness, heat, swelling, and pain are very common parameter for diagnosis of inflammation while catabolically generated edema is the only specific macroscopic sign for diagnosis. However, the presence of systematic inflammation detected by presence of various inflammatory mediators such as C-reactive protein (CRP), tumor necrosis factor-α (TNF-α), interleukin 1 (IL-1), lipoprotein-associated phospholipase A2 (Lp-PLA2) and eicosanoids with the help of sophisticated analyzing techniques.\textsuperscript{5,6}
8.1. 6. Treatment

Management of inflammation may involve steroidal anti-inflammatory drugs such as corticosteroids, Non-Steroidal Anti-Inflammatory Drugs (NSAID), Immune Selective Anti-Inflammatory Derivatives (ImSAIDs) such as FEG and SGP-T, phytochemicals and thermotherapy.\(^7,8\)

NSAIDs are most common and effective therapy for treatment of inflammation which may categorized as per following.\(^9\)

- Non selective COX inhibitor
  - Salicylates: Aspirin
  - Propionic acid derivatives: Ibuprofen, Naproxen, Ketoprofen, Flurbiprofen.
  - Fennamate: Mefennamic acid
  - Enolic acid derivatives: Piroxicam, Tenoxiam
  - Acetic acid derivatives: Ketorolac, Indomethacine, Nabumeton.
  - Pyrazole derivatives: Phenylbutazone, Oxyphenbutazone
- Preferential COX-2 inhibitors: Nimesulide, Diclofenac, Aceclofenac, Eloxican.
- Selective COX-2 Inhibitor: Celecoxib, Etoroxib, Parecoxib.

8.1. 7. Prevalence

With the advancement of molecular pathogenesis it cleared that Pro-inflammatory mediators (cytokines) have significant role in pathogenesis of various diseases like Acquired Immunodeficiency Syndrome (AIDS), asthma, cancer, cardiovascular disorders such as atherosclerosis, congestive heart failure, gastrointestinal disorders such as peptic ulcer, crohn’s disease, neurological diseases such as alzheimer’s disease, down’s syndrome, multiple sclerosis, diabetes, psychiatric disorders such as depression, schizophrenia, sleep disorders, stress, rheumatoid arthritis, sepsis.\(^10\)
However as per the survey of American cancer society cardiovascular diseases, cancer, respiratory infections, digestive diseases, and diabetes mellitus are the leading causes of death worldwide.\(^\text{11}\) Hence it can estimate that each and every person all over the world may suffer from inflammation.

### 8.1. 8. Plants having anti-inflammatory activity

Most of plants possess anti-inflammatory properties plays significant role in many inflammatory disorders while purified natural compounds from plants can serve as starting material for the synthesis of new generation anti-inflammatory drugs with low toxicity and higher therapeutic value. Some anti-inflammatory plants with their active phytochemicals are mentioned in table 8.1.\(^\text{12}\)

#### Table 8.1. List of plants having anti-inflammatory activity

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Botanical Name</th>
<th>Common Name</th>
<th>Chemical Constituent</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Acacia catechu</em></td>
<td>Katha</td>
<td>Tannins, catechin, quercetin, catechuic acid.</td>
</tr>
<tr>
<td>2</td>
<td><em>Amaranthus spinosus</em></td>
<td>Prickly amaranth</td>
<td>α-Spinasterols, octacosanoate and saponin.</td>
</tr>
<tr>
<td>3</td>
<td><em>Asystasia dalzelliana</em></td>
<td>Lavana-valli</td>
<td>Alkaloids, saponins, cardiac glycosides, flavanoids, anthraquin.</td>
</tr>
<tr>
<td>4</td>
<td><em>Butea monosperma</em></td>
<td>Palash</td>
<td>Flavonoids, chalcones, tannins.</td>
</tr>
<tr>
<td>5</td>
<td><em>Calotropis giganteas</em></td>
<td>Crown flower</td>
<td>Calotropnaphthalene, terpenes.</td>
</tr>
<tr>
<td>6</td>
<td><em>Cassia sophera</em></td>
<td>Kasunda</td>
<td>Flavonoids, glycosides</td>
</tr>
<tr>
<td>7</td>
<td><em>Cissampelos pareira</em></td>
<td>Akanadi</td>
<td>Alkaloids, flavonurine, volatile oil, quercitol.</td>
</tr>
<tr>
<td>8</td>
<td><em>Cissus quadrangularis</em></td>
<td>Hadjod</td>
<td>Flavonoids, coumarins, steroids.</td>
</tr>
<tr>
<td>No.</td>
<td>Species</td>
<td>Family</td>
<td>Common Name</td>
</tr>
<tr>
<td>-----</td>
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<td>-------------</td>
</tr>
<tr>
<td>9</td>
<td><em>Cissus rependa</em></td>
<td>Vitaceae</td>
<td>Pani bel</td>
</tr>
<tr>
<td>10</td>
<td><em>Dorstonia brasiliensis</em></td>
<td>Moraceae</td>
<td>Carapia</td>
</tr>
<tr>
<td>11</td>
<td><em>Elephantopus scaber</em></td>
<td>Asteraceae</td>
<td>Elephant foot</td>
</tr>
<tr>
<td>12</td>
<td><em>Hibiscus tiliaceus</em></td>
<td>Malvaceae</td>
<td>Beach Hibiscus</td>
</tr>
<tr>
<td>13</td>
<td><em>Holarrhena antidysenterica</em></td>
<td>Apoynaceae</td>
<td>Indrajao</td>
</tr>
<tr>
<td>14</td>
<td><em>Kaempferia galangal</em></td>
<td>Zingiberaceae</td>
<td>Aromatic ginger</td>
</tr>
<tr>
<td>15</td>
<td><em>Leucas cephalotes</em></td>
<td>Labiatae</td>
<td>Dronpushpi</td>
</tr>
<tr>
<td>16</td>
<td><em>Mangifera indica</em></td>
<td>Anarcardiaceae</td>
<td>Mango</td>
</tr>
<tr>
<td>17</td>
<td><em>Mitragyna parvifolia</em></td>
<td>Rubiaceae</td>
<td>Kaddamkamgi</td>
</tr>
<tr>
<td>18</td>
<td><em>Oxalis corniculata</em></td>
<td>Oxalidaceae</td>
<td>Creeping oxalis</td>
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<tr>
<td>19</td>
<td><em>Phyllanthus niruri</em></td>
<td>Phyllanthaceae</td>
<td>Gulf-leaf flower</td>
</tr>
<tr>
<td>20</td>
<td><em>Rubia cordifolia</em></td>
<td>Rubiaceae</td>
<td>Indian Madder</td>
</tr>
<tr>
<td>21</td>
<td><em>Solanum trilobatum</em></td>
<td>Solanaceae</td>
<td>Alarka</td>
</tr>
<tr>
<td>22</td>
<td><em>Sterculia foetida</em></td>
<td>Sterculiaceae</td>
<td>Janglibadam</td>
</tr>
<tr>
<td>23</td>
<td><em>Tectona grandis</em></td>
<td>Vervenaceae</td>
<td>Sagwan</td>
</tr>
</tbody>
</table>
8.1.9. Methods for evaluation of anti-inflammatory activity

Various numbers of methods are available for evaluation of anti-inflammatory activity which may divide into in-vitro, in-vivo evaluation for measuring acute inflammation, sub acute inflammation and chronic repair processes. The some methods were mentioned bellow.¹³

❖ In-vitro methods for anti-inflammatory activity

➢ 3H-Bradykinin receptor binding

➢ Substance P and the tachykinin family

✓ 3H-Substance P receptor binding

✓ Neurokinin receptor binding

✓ Characterization of neurokinin agonists and antagonists by biological assays

➢ Assay of polymorphonuclear leukocyte chemotaxis

➢ Polymorphonuclear leukocytes aggregation induced by FMLP

➢ Constitutive and inducible cellular arachidonic acid metabolism

✓ Formation of leukotriene B4 in human white blood cells

✓ Formation of lipoxygenase products from 14C-arachidonic acid in human polymorphonuclear neutrophils (PMN)

✓ Formation of eicosanoids from 14C-arachidonic acid in human platelets

Stimulation of inducible prostaglandin pathway in human PMNL

✓ COX-1 and COX-2 inhibition

➢ Induced release of cytokines (Interleukin-1alpha, IL-1beta, IL-6, IL-8 and TNF-alpha) from human white blood cells in vitro

➢ Flow cytometric analysis of intracellular cytokines

➢ TNF-alpha antagonism
Anti-inflammatory Evaluation of Meyna laxiflora

- Binding to interferon receptors
- Screening for interleukin-1 antagonists
- Inhibition of interleukin-1β converting enzyme (ICE)

❖ In-vivo methods for anti-inflammatory activity

- Methods for testing acute and subacute inflammation
  - Ultraviolet erythema in guinea pigs
  - Vascular permeability
  - Inhibition of leukocyte adhesion to rat mesenteric venules
  - Oxazolone-induced ear edema in mice
  - Croton-oil ear edema in rats and mice
  - Paw edema
  - Pleurisy test
  - Granuloma pouch technique
  - Urate-induced synovitis

- Methods for testing the proliferative phase (granuloma formation)
  - Cotton wool granuloma
  - Sponge implantation technique
  - Glass rod granuloma
8.2. Methodology

Anti-inflammatory activity was evaluated at Deshpande Laboratories Pvt. Ltd. Bhopal, with prior permission of CPCSEA/IAEC (DL/MA/ 11/13/102) by determining formalin induced rat hind paw edema and carrageenan induced rat hind paw edema.

❖ Preparation of solutions

The test samples (petroleum ether, chloroform, methanolic and aqueous extracts of plant) and standard drug (Diclofenac) were prepared as a suspension in distilled water using mortar and pestle.

❖ Selection of dose

Earlier studies on plants showed that various extracts of plant leaves did not produce any mortality at the dose of 2000 mg/kg thus considered as approximate medium lethal dose (ALD\textsubscript{50}) which comes under category 5 (safe dose) under Globally Harmonized Classification System (GHS).\textsuperscript{14} Hence a dose of 100, 200 and 400 mg/kg body weight were chosen for the study.

❖ Selection of animals and preparation of groups

The Wistar rats weighing 150 to 200g were selected for the study and divided into fifteen groups, each groups contain three rats. All the rats were housed in standard plastic rat cages with stainless steel coverlids with standard environmental condition of photoperiod (12:12 h dark: light cycle) and temperature (25 ± 2°C). Animals were fed standard pellet diet and water ad libitum.
**Group I:** Untreated & Un-induced

**Group II:** Untreated

**Group III:** Diclofenac 10mg/kg

**Group IV (MLA 100 mg/kg):** Petroleum ether extract 100 mg/kg

**Group V (MLA 200 mg/kg):** Petroleum ether extract 200 mg/kg

**Group VI (MLA 400 mg/kg):** Petroleum ether extract 400 mg/kg

**Group VII (MLB 100 mg/kg):** Chloroform extract 100 mg/kg

**Group VIII (MLB 200 mg/kg):** Chloroform extract 200 mg/kg

**Group IX (MLB 400 mg/kg):** Chloroform extract 400 mg/kg

**Group X (MLC 100 mg/kg):** Methanolic extract 100 mg/kg

**Group XI (MLC 200 mg/kg):** Methanolic extract 200 mg/kg

**Group XII (MLC 400 mg/kg):** Methanolic extract 400 mg/kg

**Group XIII (MLD 100 mg/kg):** Aqueous extract 100 mg/kg

**Group XIV (MLD 200 mg/kg):** Aqueous extract 200 mg/kg

**Group XV (MLD 400 mg/kg):** Aqueous extract 400 mg/kg

❖ **Carrageenan induced hind paw edema**

After grouping, the animals initial paw volume was measured using vernier calipers (Mitutoyo, JAPAN). Thereafter, diclofenac 10mg/kg or test samples at 100, 200, 400mg/kg or distilled water were orally administered to the respective group of the animals. 2 hours post drug administration, 0.1 ml of carrageenan was injected in the right hind leg of the animal, edema formed in the paw was measured by digital vernier calipers after 3 hours. The degree of swelling induced was evaluated by the ratio of the volume of hind paw before to after carrageenan treatment. The percentage inhibition was determined by considering edema induced by carrageenan alone was as 100% induction. Results are expressed as mean ± SEM. The statistical analysis
was performed by one way analyses of variance (ANOVA) followed by Bonferroni multiple comparisons test with GraphPad Istant3. The $P<0.05$ was considered as statistically significant.

**Formalin induced hind paw edema**

After grouping, the animals initial paw volume was measured using vernier calipers (Mitutoyo, JAPAN). Thereafter, diclofenac 10mg/kg or test samples at 100, 200, 400mg/kg or distilled water were orally administered to the respective group of the animals. 2 hours post drug administration, 0.1 ml of 2% Formalin was injected in the right hind leg of the animal, edema formed in the paw was measured by digital vernier calipers after 3 hours. The degree of swelling induced was evaluated by the ratio of the volume of hind paw before to after Formalin treatment. The percentage inhibition was determined by considering edema induced by Formalin alone was as 100% induction. Results are expressed as mean ± SEM. The statistical analysis was performed by one way analyses of variance (ANOVA) followed by Bonferroni multiple comparisons test with GraphPad Istant3. The $P<0.05$ was considered as statistically significant.

### 8.3. Results and Discussion

The results of the present study showed that the all extracts of plant have the dose-dependent anti-inflammatory effects against paw edema induced by carrageenan and formalin. The results also showed that Petroleum ether extract have significant anti-inflammatory activity as compared to other extracts.
Carrageenan induced hind paw edema

The results of the present study showed that the all extracts of plant showed decrease in paw volume in carrageenan induced inflammation in rats at different doses (100 mg/kg, 200 mg/kg, 400 mg/kg). The results also showed that petroleum ether extract have significant anti-inflammatory activity as compare to other extracts. (Table 8.2) (Figure 8.4 and 8.5)

Petroleum ether extract showed percent inhibition of carrageenan induced inflammation in rats 17.83, 38.83 and 67.32 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having significant anti-inflammatory activity as compare to standard diclofenac which have 77.59 percent inhibition of inflammation at 10mg/kg.

Chloroform extract showed percent inhibition of carrageenan induced inflammation in rats 15.97, 19.7 and 22.5 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having considerable anti-inflammatory activity.

Methanolic extract showed percent inhibition of carrageenan induced inflammation in rats 10.36, 16.90 and 24.37 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having considerable anti-inflammatory activity.

Aqueous extract showed percent inhibition of carrageenan induced inflammation in rats 17.83, 25.3 and 44.91 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having significant anti-inflammatory activity at 400 mg/kg dose.
Table 8.2. Effect of different extracts of *Meyna laxiflora* leaf on carrageenan induced hind paw edema

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Animal groups</th>
<th>Degree of swelling</th>
<th>% Inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Untreated &amp; Un-induced</td>
<td>1.00 ± 0.00</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>Untreated</td>
<td>1.57 ± 0.01</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>Diclofenac 10mg/kg</td>
<td>1.13 ± 0.03</td>
<td>77.59</td>
</tr>
<tr>
<td>4</td>
<td>(MLA 100 mg/kg)</td>
<td>1.49 ± 0.02</td>
<td>17.83</td>
</tr>
<tr>
<td>5</td>
<td>(MLA 200 mg/kg)</td>
<td>1.36 ± 0.04</td>
<td>38.38</td>
</tr>
<tr>
<td>6</td>
<td>(MLA 400 mg/kg)</td>
<td>1.19 ± 0.05</td>
<td>67.32</td>
</tr>
<tr>
<td>7</td>
<td>(MLB 100 mg/kg)</td>
<td>1.50 ± 0.07</td>
<td>15.97</td>
</tr>
<tr>
<td>8</td>
<td>(MLB 200 mg/kg)</td>
<td>1.47 ± 0.06</td>
<td>19.7</td>
</tr>
<tr>
<td>9</td>
<td>(MLB 400 mg/kg)</td>
<td>1.46 ± 0.04</td>
<td>22.5</td>
</tr>
<tr>
<td>10</td>
<td>(MLC 100 mg/kg)</td>
<td>1.55 ± 0.02</td>
<td>10.36</td>
</tr>
<tr>
<td>11</td>
<td>(MLC 200 mg/kg)</td>
<td>1.48 ± 0.02</td>
<td>16.9</td>
</tr>
<tr>
<td>12</td>
<td>(MLC 400 mg/kg)</td>
<td>1.44 ± 0.02</td>
<td>24.37</td>
</tr>
<tr>
<td>13</td>
<td>(MLD 100 mg/kg)</td>
<td>1.48 ± 0.02</td>
<td>17.83</td>
</tr>
<tr>
<td>14</td>
<td>(MLD 200 mg/kg)</td>
<td>1.46 ± 0.02</td>
<td>25.3</td>
</tr>
<tr>
<td>15</td>
<td>(MLD 400 mg/kg)</td>
<td>1.32 ± 0.03</td>
<td>44.91</td>
</tr>
</tbody>
</table>

Data are mean of 3 replicates ± SEM and are significantly different at *P* < 0.05.
Anti-inflammatory Evaluation of *Meyna laxiflora*

**Figure 8.4.** Degree of swelling of different extracts of *Meyna laxiflora* leaf in carrageenan induced hind paw edema

**Figure 8.5.** Percent inhibition of different extracts of *Meyna laxiflora* leaf in carrageenan induced hind paw edema
Formalin induced hind paw edema

The results of the present study showed that the all extracts of plant showed decrease in paw volume in formalin induced inflammation in rats at different doses (100 mg/kg, 200 mg/kg, 400 mg/kg). The results also showed that petroleum ether extract have significant anti-inflammatory activity as compare to other extracts. (Table 8.3) (Figure 8.6 and 8.7)

Petroleum ether extract showed percent inhibition of formalin induced inflammation in rats 14.07, 27.29 and 49.01 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having significant anti-inflammatory activity as compare to standard diclofenac which have 62.23 percent inhibition of inflammation at 10mg/kg.

Chloroform extract showed percent inhibition of formalin induced inflammation in rats 13.13, 15.96 and 16.90 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having considerable anti-inflammatory activity.

Methanolic extract showed percent inhibition of formalin induced inflammation in rats 7.46, 13.13 and 25.40 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having considerable anti-inflammatory activity.

Aqueous extract showed percent inhibition of formalin induced inflammation in rats 12.18, 17.85 and 30.12 at 100, 200 and 400 mg/kg dose respectively after three hours which indicates that it having considerable anti-inflammatory activity.
Table 8.3. Effect of different extracts of *Meyna laxiflora* leaf on formalin induced hind paw edema

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Animal groups</th>
<th>Degree of swelling</th>
<th>% Inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Untreated &amp; Un-induced</td>
<td>1.00 ± 0.00</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>Untreated</td>
<td>1.57 ± 0.02</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>Diclofenac 10mg/kg</td>
<td>1.22 ± 0.06</td>
<td>62.23</td>
</tr>
<tr>
<td>4</td>
<td>(MLA 100 mg/kg)</td>
<td>1.50 ± 0.02</td>
<td>14.07</td>
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<tr>
<td>5</td>
<td>(MLA 200 mg/kg)</td>
<td>1.42 ± 0.05</td>
<td>27.29</td>
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<td>6</td>
<td>(MLA 400 mg/kg)</td>
<td>1.29 ± 0.02</td>
<td>49.01</td>
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<tr>
<td>7</td>
<td>(MLB 100 mg/kg)</td>
<td>1.49 ± 0.02</td>
<td>13.13</td>
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<td>8</td>
<td>(MLB 200 mg/kg)</td>
<td>1.48 ± 0.02</td>
<td>15.96</td>
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<td>9</td>
<td>(MLB 400 mg/kg)</td>
<td>1.47 ± 0.01</td>
<td>16.9</td>
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<tr>
<td>10</td>
<td>(MLC 100 mg/kg)</td>
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<tr>
<td>11</td>
<td>(MLC 200 mg/kg)</td>
<td>1.49 ± 0.04</td>
<td>13.13</td>
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<tr>
<td>12</td>
<td>(MLC 400 mg/kg)</td>
<td>1.42 ± 0.01</td>
<td>25.4</td>
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<tr>
<td>13</td>
<td>(MLD 100 mg/kg)</td>
<td>1.50 ± 0.03</td>
<td>12.18</td>
</tr>
<tr>
<td>14</td>
<td>(MLD 200 mg/kg)</td>
<td>1.47 ± 0.02</td>
<td>17.85</td>
</tr>
<tr>
<td>15</td>
<td>(MLD 400 mg/kg)</td>
<td>1.40 ± 0.03</td>
<td>30.12</td>
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</table>

Data are mean of 3 replicates ± SEM and are significantly different at *P* < 0.05.
Figure 8.6. Degree of swelling of different extracts of *Meyna laxiflora* leaf in formalin induced hind paw edema

Figure 8.7. Percent inhibition of different extracts of *Meyna laxiflora* leaf in formalin induced hind paw edema
From the above result it is cleared that petroleum ether extract have significant anti-inflammatory activity. The quantitative phytochemical analysis of *Meyna laxiflora* reveals that petroleum ether extracts contain high concentration of steroids and saponin as compare to other.

Anti-oxidants such as vitamins, carotenoids, tannin, flavonoids, phenolic compounds and steroids responsible for anti-inflammatory activity considering that most of inflammation reaction due to reactive oxygen and nitrogen species activates monocytes and macrophages for production of pro-inflammatory cytokine by depleting intracellular thiol compounds and activating nuclear factor KB (NF-KB).\textsuperscript{7,17} Hence, Hence it can be concluded that high concentrations of steroids in petroleum ether extracts may responsible for significant anti-inflammatory activity.

8.4. References


