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***In-vivo* and *in-vitro* screening of medicinal plants for their anti-inflammatory activity: an overview**

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ABSTRACT

Despite the progress made in medical research for the past decades, the treatment of many serious diseases is still problematic. Inflammation is the response of living tissues to injury. It involves a complex array of enzyme activation, mediator release, extravasations of fluid, cell migration, tissue breakdown and repair. Inflammation has become the focus of global scientific research because of its implication in virtually all human and animal diseases. As a result of adverse effects such as gastric lesions caused by non-steroidal anti-inflammatory drugs (NSAID), tolerance and dependence induced by opiates, the use of these drugs as anti-inflammatory agents have not been successful in all cases. Therefore, new anti-inflammatory drugs lacking these side effects are being researched as alternatives to NSAID and opiates. Attention is being focused on the investigation of the efficacy of plant-based drugs used in the traditional medicine because they are cheap, have little side effects. Hence, in the present review the various animal models used for preclinical screening anti-inflammatory activity herbs was compiled.

Keywords: Inflammation, Animal Models, Plant Extract, Mechanism of action.

INTRODUCTION

Inflammation is part of the complex biological response of vascular tissues to harmful stimuli, such as pathogens, damaged cells, or irritants. Inflammation is a protective attempt by the organism to remove the injurious stimuli and to initiate the healing process. Inflammation is not a synonym for infection, even in cases where inflammation is caused by infection. Although infection is caused by a microorganism, inflammation is one of the responses of the organism to the pathogen. Without inflammation, wounds and infections would never heal. Similarly, progressive destruction of the tissue would compromise the survival of the organism (Wikipedia). The plants are one of the most important sources of medicines. India is known due to availability of several thousands of medicinal plants in the different bioclimatic zones anti-inflammatory diseases (Hemamalini *et al.*, 2010).

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DIFFERENT MODELS USED TO STUDY INFLAMMATION

Acetic acid induced vascular permeability (Lee *et al.*, 2009).

Plant used: *Asparagus cochinchinensis*.

Family: Liliaceae.

Part used: Dried and pulverized roots.

Common name: Asparagus root, shiny asparagus.

Solvent: 70% ethanol.

Yield: Not mentioned.

Dose: 2 g/kg.

Route of administration: Orally

Chemical used: Indomethacin, Evans blue dye, Acetic acid.

Procedure

Acetic acid-induced vascular permeability in ICR mice was performed as described previously. Briefly, 1 h after oral administration of ACE (200mg/kg), indomethacin (5 mg/kg), or an equivalent volume of vehicle (3%, v/v Tween 80), 0.2 ml of Evans blue dye (0.25% in normal saline) was administered intravenously through the tail vein. Thirty minutes later, the animals received 1 ml/100 g of acetic acid (0.6%, v/v) i.p. Treated animals were sacrificed 30min after acetic acid injection and the peritoneal cavity washed with normal saline (3 ml) into heparinized tubes and centrifuged. The dye content of the supernatant was measured at 610 nm using a microplate reader (molecular device). The results were expressed as arbitrary units of relative value.

Mechanism of action

The anti-inflammatory activity of *A. cochinchinensis* with 70% ethanol extract may be due to blockade of pro-inflammatory cytokine production, neutrophil-mediated myeloperoxidase activity, inhibition of IL-1 β and TNF- α and to the subsequent blockade of leukocyte accumulation

Conclusion

These results demonstrate that ACE is an effective anti-inflammatory agent in murine phorbol ester-induced dermatitis, and suggest that the compound may have therapeutic potential in a variety of immune-related cutaneous diseases.

Carrageenan – induced oedema.(Ilavarasan *et al.*, 2005).

Plant used:- *Cassia fistula* Linn.

Family:- Caesalpinaceae.

Part used:- Bark.

Common name:- Amalltas.

Solvent:- Water and methanol.

Yield:- Water- 7% w/w and methanol- 9% w/w.

Dose:- 2000 mg/kg.

Route of administration:- Orally.

Chemical used:- Diclofenac sodium.

Procedure

Paw oedema was induced by injecting 0.1ml of 1% Carrageenan in physiological saline into the subplantar tissues of

the left hind paw of each rat. The extracts CFA (*C. fistula* Aqueous), CFM (*C.fistula* Methanolic) were administered orally 30 min prior to Carrageenan administration. The paw volume was measured at 60, 120, 180, 240, minutes by the mercury displacement method using a plethysmograph. The percentage inhibition of paw volume in drug treated group was compared with the control group. Diclofenac sodium was used as reference standard.

Mechanism of action

The activity of *C. fistula* Linn. with aqueous and methanolic extracts may be due to inhibition of the mediators of inflammation such as histamine, serotonin and prostaglandin.

Conclusion

Plants which belong to *Caesalpinaceae* family are rich in flavonoids and bio flavonoids are known for their anti-inflammatory and antioxidant activities. Further research is in progress to identify the biomolecules responsible for the anti-inflammatory and antioxidant activities.

Carrageenan induced air pouch model (Paschapur *et al.*, 2009.)

Plant used:- *Borassus flabellifer* Linn.

Family:- Arecaceae.

Part used:- Male flowers.

Common name:- Palmyra palm, toddy or wine palm.

Solvent:- Ethanol.

Yield:- Not mentioned.

Dose:- 150 and 300mg/kg.

Route of administration:- Orally.

Chemical used:- Pure Diclofenac Sodium.

Procedure

The rats were divided into four groups. Rats were anesthetized and air cavities were produced by subcutaneous injection of 20 ml of sterile air into the intrascapular area of the back. An additional 10 ml of air was injected into the cavity every 3rd day to keep the space open. On the 7th day, 2 ml of 1% solution of carrageenan dissolved in saline was injected directly into the pouch to induced an inflammatory response. The rats were orally pre-treated with either vehicle or extract or diclofenac sodium 3 h prior to the injection of carrageenan. The second dose of treatment was repeated after 24 h of the first treatment. 48 h after carrageenan injection, the rats were anesthetized with ether and the pouch was carefully opened by a small incision. The volume of exudates was collected and measured. An aliquot of the exudate was used for quantification of leukocyte concentration using a haemocytometer and differential cell count was performed using a manual cell counter after staining with Wright's stain. The results were expressed as the total number of neutrophils and monocytes.

Mechanism of action

The anti-inflammatory activity of *B. flabellifer* Linn. may be due to inhibiting either release of lysosomal enzymes or by stabilizing the lysosomal membrane, which is one of the major events responsible for the inflammatory process.

Conclusion

Plants which belong to Caesalpinaceae family are rich in flavonoids and bio flavonoids are known for their anti-inflammatory and antioxidant activities. Further research is in progress to identify the biomolecules responsible for the anti-inflammatory and antioxidant activities.

Carrageenan-induced pleurisy in rats (Badilla *et al.*, 2003.).

Plant used: *Loasa speciosa*.

Family: Loasaceae.

Part used: Leaves.

Common name: Campana.

Solvent: Water.

Yield: Not mentioned.

Dose: 250–500 mg/kg.

Route of administration: Intraperitoneally.

Chemical used: Indomethacin.

Procedure

Groups of six rats each were pretreated with the test extract (250 or 500 mg/kg, i.p.), indomethacin (10 mg/kg, i.p.) or saline (0.1 ml). One hour later all animals received an intrapleural injection of carrageenan (0.25 ml of 1% solution in 0.9% saline) on the right side of the thorax. Three hours later the animals were anaesthetized with ether and killed by bleeding through the portal vein. The pleural exudate was collected and the pleural cavity was washed with 1.0-ml saline containing heparin (10 IU/ml). The number of migrating leukocytes in the exudate was determined with a Neubauer chamber. Results were expressed as mean \pm S.E.M. of exudate pleural volumes (ml) and of total leukocytes.

Mechanism of action

The aqueous extract of *L. speciosa* leaves showed an inhibitory effect on leukocyte migration, and a reduction on the pleural exudates.

Conclusion

When given intraperitoneally, has analgesic and anti-inflammatory activities, which are dose-dependent over a range of 125–500 mg/kg. Further studies are necessary to assess the potential clinical use of this plant, or its extract or active principles, as anti-inflammatories and or analgesics.

Carrageenin- and arachidonic acid-induced paw edema

(Ghule *et al.*, 2006.).

Plant used: *Lagenaria siceraria*.

Family: Cucurbitaceae.

Part used: Fresh fruits.

Common name: Bottle gourd.

Solvent: Stand. fruit juice.

Yield: 22.5 % w/w.

Dose: 300 mg/kg.

Route of administration: Orally.

Chemical used: Arachidonic acid.

Procedure

Male rats weighing between 100–150 g were used. Paw edema was induced by an intradermal injection of carrageenin (1% in normal saline solution) or arachidonic acid (0.5% in 0.2 M carbonate buffer, pH 8.4) into the plantar surface of the right hind paw of the rats, at a volume of 0.05 or 0.1 ml, respectively. The edema volume was determined using a plethysmometer prior to and 1, 3 and 5 h after carrageenin injection, or 1 h after arachidonic acid injection. Test drugs were given 1 h prior to carrageenin or 2 h prior to arachidonic acid injection.

Mechanism of action

The fruit juice extract of *L. siceraria* has a good predictive value to screen anti-inflammatory agents. Inflammatory mediators such as kinin, serotonin, and PGs are released by EPP. It was found that the LSFJE elicited significantly inhibitory effect on the edema formation at all assessment times. Phenylbutazone, a COX-inhibitor, showed a marked reduction of the ear edema.

Conclusion

The majority of its activities appear to involve or depend on arachidonic acid release and metabolism and interaction with protein kinase C. It has a good predictive value to screen anti-inflammatory agents. Inflammatory mediators such as kinin, serotonin, and PGs are released by EPP.

Cotton pellet induced granuloma (Ilavarasan *et al.*, 2005.).

Plant used: *Cassia fistula* Linn.

Family: Caesalpinaceae.

Part used: Bark.

Common name: Amalltas.

Solvent: Water and methanol.

Yield: Water- 7% w/w and methanol- 9% w/w.

Dose: 2000 mg/kg.

Route of administration: Orally.

Chemical used: Diclofenac sodium.

Procedure

Wistar albino rats (170–200 gm) of either sex were divided into 4 groups of 6 animals in each group. Cotton pellets weighing 30 \pm 1 mg were autoclaved and implanted subcutaneously into both sides of the groin region of each rat. Group I served as control and received the vehicle. The Extracts CFA and CFM at concentration 500 mg/kg orally for Group II and III animals for same period. On the 8th day the animals were sacrificed and the pellets together with the granuloma tissues were carefully removed, dried in an oven at 60°C, weighed and compared with control. Diclofenac sodium (5 mg / kg / p.o.) was used as reference standard.

Mechanism of action

The anti-inflammatory activity of *C. fistula* Linn. With aqueous and methanolic extracts may be due to inhibition of the mediators of inflammation such as histamine, serotonin and prostaglandin.

Conclusion

Plants which belong to *Caesalpinaceae* family are rich in flavonoids and bio flavonoids are known for their anti-inflammatory and antioxidant activities.

Croton oil-induced mouse ear edema (Canigual *et al.*, 1998.).

Plant used: *Anthurium cerrocampanense*.

Family: Araceae.

Part used: Whole plant.

Common name: Tailflower, anthrium, flamingo flower.

Solvent: Water, Ethanol, dichloromethane.

Yield: Dichloromethane-1%, ethanol- 1.34%, Water-4.7%.

Dose: 100mg/kg.

Route of administration: Intraperitoneally.

Chemical used: Indomethacin.

Procedure

Groups of 10 animals were formed. Each animal was anesthetized with ketamine. After 5 min, each animals received 15 μ l of a solution of croton oil in acetone or ethanol 43%, on the inner surface of the right ear. The extracts and reference drug were dissolved in the solution of croton oil. Five and a half hours after the administration of croton oil, the animals were sacrificed by cervical dislocation, and a 7 mm diameter disc from each ear was removed with a stainless steel punch. The edema size was determined in relation to the weight of untreated left ear.

Mechanism of action

The inflammatory process is caused by the release of mediators from tissues and migrating cells, and most strongly implicated are the prostaglandines (PGs), leucotrienes (LTs), histamine, bradykinin, platelet-activating factor (PAF) and interleukin-1.

Conclusion

activity is related to histamine and serotonin mediators (Nishida *et al.*, 1979; Nishida and Tomizawa, 1980). It is known that on arachidonic acid-induced rat paw edema, araquidonic acid derivatives, specially leucotriens, have an important role and the COX inhibitors show low or no activity.

Cutaneous inflammation induced to the inner surface of right ear (Manga *et al.*, 2004.).

Plant used: *Alchornea cordifolia*.

Family: Euphorbiaceae.

Part used: Leaves.

Common name: Christmas bush.

Solvent: Methanol.

Yield: 13.08% w/w.

Dose: 2000 μ g/cm².

Route if administration: Topically.

Chemical used: Indomethacin.

Procedure

Cutaneous inflammation was induced to the inner surface of the right ear (surface: about 1 cm²) of anaesthetized mice (145 mg/kg ketamine hydrochloride, intraperitoneally) by application of 15 μ l of acetone or acetone–water (1:1) solutions containing 80 μ g of Croton oil as irritant (control animals). For treated animals, appropriate amounts of indomethacin (reference drug) or of the tested extracts were dissolved in the Croton oil containing solution and applied as for controls. After 6 h, mice were sacrificed and a punch (5mm of diameter) was excised from both ears. Inflammation was measured as oedema formation and quantified by the weight difference between the treated and the untreated (opposite) ear samples.

Mechanism of action

The anti inflammatory activity of methanolic extract of *A. cordifolia* may reduced the inflammation in to the inner surface of the right ear.

Conclusion

The Methanol extracts of *A. cordifolia* leaves showed strong dose-dependent anti-inflammatory activity after topical application. This activity may be explained at least partially by unidentified lipophilic compounds and by flavonoids (e.g. hyperoside and quercitrin) but the degree of activity was not related to tannin contents.

Egg white induced hind paw edema (Arunachalam *et al.*, 2009.).

Plant used: *Eclipta prostrata* L.

Family: Astearaceae.

Part used: Leaves.

Common name: Trailing, Bhamgra.

Solvent: Methanol.

Yield: 8.7% w/w.

Dose: 100-200 mg/kg.

Route of administration: Orally.

Chemical used: Propylene glycol.

Procedure

Albino Wistar rats of either sex weighing about 160 - 180 g were divided into four groups of six animals each. The methanol extract of leaves of *E. prostrata* at 100 and 200 mgkg⁻¹ was administered orally to first two groups of rats. The third and fourth group of rats received 5 mlkg⁻¹ propylene glycol as vehicle control or 8 mgkg⁻¹ cyproheptadine as drug control respectively, for comparative pharmacological assessment.

All the drugs and vehicle were given 1 h prior to the study. Freshly taken egg white (0.1 ml) was injected into the sub plantar tissue of the left hind paw of the rat. The volumes of the injected paws were measured at 0, 60, 120, 180 and 240 min using a plethysmometer. The percent increase in paw oedema of the treated group was compared with that of the control and the inhibitory effects of the drugs were studied.

Mechanism of action

The anti-inflammatory activity of methanolic extract of *E. prostrata* L. has an inhibitory effect on the release of active pain substance such as histamine, serotonin, polypeptides or prostaglandins.

Conclusion

The preliminary phytochemical screening of leaves of *E. prostrata* indicated the presence of steroids, triterpenoids, flavanoids, tannins, reducing sugar and saponins. The steroids, alkaloids and triterpenoids present in the extract may be responsible for this anti-oedematous effect.

Ethyl phenylpropionate-induced ear edema

(Ghule *et al.*, 2006.).

Plant used: *Lagenaria siceraria*.

Family: Cucurbitaceae.

Part used: Fresh fruits.

Common name: Bottle gourd.

Solvent: Stand. fruit juice.

Yield: 22.5 % w/w.

Dose: 300 mg/kg.

Route of administration: Orally.

Chemical used: Ethyl phenylpropionate.

Procedure

Male rats weighing 100-150 g were used. Ear edema was induced by topical application of EPP at a dose of 1 mg/20 µl per ear to the inner and outer surfaces of both ears by means of an automatic microliter pipette. Test drugs were applied topically in volumes of 20 µl just before the irritant. The control group received vehicle only. Before and at 30min, 1h and 2h after edema induction, the thickness of each ear was measured by vernier calipers. The percent inhibition of the edema formation of test substances was calculated.

Mechanism of action

The inflammatory activity appear to involve or depend on arachidonic acid release and metabolism and interaction with protein kinase. The fruit juice extract of *L. siceraria* has a good predictive value to screen anti-inflammatory agents. Inflammatory mediators such as kinin, serotonin, and PGs are released by EPP. It was found that the LSFJE elicited significantly inhibitory effect on the edema formation at all assessment times. Phenylbutazone, a COX-inhibitor, showed a marked reduction of the ear edema.

Conclusion

EPP-induced ear edema test provides a skin inflammation model suitable for evaluation of topical and systemic anti-inflammatory agents. The majority of its activities appear to involve or depend on arachidonic acid release and metabolism and interaction with protein kinase C.

Formalin induced edema in rat paw (Hosseinzadeh., 2002.).

Plant used: *Crocus sativus* Linn.

Family: Iridaceae.

Part used: Powder of stigma and petal.

Common name: Saffron.

Solvent: Water and ethanol.

Yield: For water-50.8%w/w and ethanol- 56.6%w/w and for petal, water-15.5% w/w and ethanol- 19%w/w.

Dose: 0.8 g/kg and 2 g/kg.

Route of administration: Intraperitoneally.

Chemical used: Formaldehyde, Diclofenac.

Procedure

Rats were divided into groups of six. The inflammation was produced by subponeurotic injection of 0.1 ml of 2 % formaldehyde in the right hind paw of the first and third day. The animals were treated daily with the extracts or diclofenac intraperitoneally for 10 days. The daily changes in paw size were measured by wrapping a piece of cotton thread round the paw and measuring the circumference with a meter rule.

Mechanism of action

The petal extracts showed anti-inflammatory activity in the chronic inflammation. The anti-inflammatory activity of the petal extracts may be due to their content of flavonoids, tannins and anthocyanins which inhibites the cyclooxygenase activity.

Conclusion

We conclude that aqueous and ethanolic extracts of saffron stigma and petal have an antinociceptive effect, as well as acute and/or chronic anti-inflammatory activity.

Formalin-induced nociception in mice (Badilla *et al.*, 2003).

Plant used: *Loasa speciosa*.

Family: Loasaceae.

Part used: Leaves.

Common name: Campana.

Solvent: Water.

Yield: Not mentioned.

Dose: 250–500 mg/kg.

Route of administration: Intraperitoneally.

Chemical used: Indomethacin.

Procedure

Each group of six mice each, was treated with the test extract (125, 250 and 500 mg/kg i.p. in saline solution), saline solution (i.p.) and indomethacin (10 mg/kg i.p.). One hour after treatment all animals were injected with 20 ml of 1% formalin in saline solution into the plantar surface of the left hind paw. Licking time was measured in the test chamber over 30 min, in two phases: the first phase was from time zero to 5 min after formalin injection

and the second phase was from 20 to 30 min after formalin injection.

Mechanism of action

The aqueous extract of *L. speciosa* leaves showed an inhibitory effect on leukocyte migration, and a reduction on the pleural exudates.

Conclusion

When given intraperitoneally, has analgesic and anti-inflammatory activities, which are dose-dependent over a range of 125–500 mg/kg. Further studies are necessary to assess the potential clinical use of this plant, or its extract or active principles, as anti-inflammatories and or analgesics.

Granuloma air pouch (Ching., 2009).

Plant used: *Stereospermum kunthianum*.

Family: Bignoniaceae.

Part used: Fresh stem bark

Common name: Sansami, umana and alakiriti.

Solvent: Water.

Yield: 26.4 % w/w.

Dose: 500 mg/kg.

Route of administration: Intraperitoneally.

Chemical used: Indomethacin.

Procedure

Wistar rats of either sex were randomly allocated into groups of five animals each. The dorsal skin surface of the rats was shaved and disinfected with 70% ethanol. Twenty millilitres of air was injected subcutaneously under ether anaesthesia at approximately the midpoint of the animals' back using a hypodermic syringe with the needle directed towards the scapular region. The animals were administered distilled water (5 ml/kg, p.o.), extract (400 mg/kg, p.o.), or indomethacin (10 mg/kg, s.c.). One hour later for the extract and thirty minutes for indomethacin treated animals respectively, 0.5 ml of carrageenan 10% w/v in olive oil was injected into the air pouch of each rat. Remnant air was removed from the air pouch 24 h after the injection of the carrageenan. The rats were treated daily with the same dose of extract or indomethacin for five days with the last dose on the fifth day given one hour prior to the time the animals were sacrificed. The animals were sacrificed by inhalation of excess chloroform. The air pouch was carefully opened and the volume of exudates formed in each animal was measured.

Mechanism of action

The aqueous extract of *S. kunthianum* stem bark possesses anti-inflammatory activity which is probably related to the inhibition of prostaglandin synthesis.

Conclusion

The aqueous extracts of *S. kunthianum* stem bark possesses anti-inflammatory activity, which is probably related to the inhibition of prostaglandin synthesis.

Histamine- induced hind paw oedema (Amresh 2007).

Plant used: *Fumaria indica*.

Family: Fumariaceae.

Part used: Whole plant.

Common name: Fumitory, pitpapra, khetpapra, parpata.

Solvent: 50% ethanol.

Yield: Not mentioned.

Dose: 400 mg/ kg.

Route of administration: Orally.

Chemical used: Phenylbutazone.

Procedure

Right hind paw oedema was induced by the sub plantar injection of 0.1 ml of histamine. Extract and phenylbutazone were administered 1 h prior to the inflammatory insult. The paw volume compared to that negative control animal was recorded after 3 h and considered as anti-inflammatory response.

Mechanism of action

The caffeic acid and b-sitosterol may be responsible for the anti-inflammatory effects exhibited by *F. indica*. The ability of the *F. indica* extract in inflammatory activity may be due to the involvement of prostaglandins and other mediators in a different order of magnitudes.

Conclusion

Based on the present study, it can be concluded that *F. indica* has potential anti-inflammatory activity against both exudative and proliferative phases of inflammation. *F. indica* extract significantly raised the pain threshold. This offers a new perspective in the treatment of pain.

Hot scald-induced rat paw edema (Yin *et al.*, 2000).

Plant used:- *Boschnikia rossica*.

Family:- Betulaceae.

Part used:- Whole plant.

Common name:- Bu Lao Cao.

Solvent:- Methanol extract was fractionated with CH₂Cl₂ and H₂O. **Yield:-** Not mentioned.

Dose:- 500-1000 mg/kg.

Route of administration:- Orally.

Chemical used:- BRH₂O extract.

Procedure

Edema was induced on the right hind paw of rat by hot scald. The right hind paw of rat soaked in thermostate water bath maintained at 53 ± 0.5 °C and cut-off time was 14 sec and test drug (500mg/kg of BRH₂O extract) was given 30 minutes before the hot scald test. The volume of the right paw was measured before test and 1, 2, 3, 4, 5, 6 and 24 h after induction of inflammation.

Mechanism of action

B. rossica extract exhibited inhibitory effect on formation of preneoplastic hepatic foci in early stage of rat chemical

hepatocarcinogenesis. Both CH₂Cl₂ and H₂O extracts from *B. rossica* exerted anti-inflammatory effect in rats and mice.

Conclusion

B. rossica extract exhibited inhibitory effect on formation of preneoplastic hepatic foci in early stage of rat chemical hepatocarcinogenesis. Both CH₂Cl₂ and H₂O extracts from BR exerted anti-inflammatory effect in rats and mice.

HRBC membrane stabilization (Jain *et al.*, 2009).

Plant used:- *Abutilon indicum* Linn.

Family: - Malvaceae.

Part used :- Leaves.

Common name: - Atibala.

Solvent: - Ethanol, chloroform and water.

Yield: - Ethanol- 4.5% w/w, chloroform- 0.45% w/w and distilled water-3.7% w/w

Dose:- 50 mg/100ml.

Route of administration:- Not mentioned.

Chemical used:- Diclophenac, phosphate buffer, HRBC suspension.

Procedure

The HRBC membrane stabilization has been used as method to study the anti-inflammatory activity. Blood was collected from healthy volunteer. The collected blood was mixed with equal volume of sterilised Alsever solution (2% dextrose, 0.8% sodium citrate, 0.5% citric acid and 0.42% sodium chloride in water). The blood was centrifuged at 3000 rpm and packed cell were washed with isosaline (0.85%, pH 7.2) and a 10% (v/v) suspension was made with isosaline. The assay mixture contained the drug. 1 ml of phosphate buffer (0.15M, pH 7.4), 2ml of hyposaline (0.36%) and 0.5 ml of HRBC suspension. Diclophenac was used as reference drug. Instead of hyposaline 2 ml of distilled water was used in the control. All the assay mixture were incubated at 37°C for 30 min and centrifuged. The hemoglobin content in the supernatant solution was estimated using spectrophotometer at 560 nm. The percentage hemolysis was calculated by assuming the hemolysis produced in presence of distilled water of as 100%.

Mechanism of action

HRBC membrane similar to lysosomal membrane components, the prevention of hypotonicity induced HRBC membrane lysis is taken as a measure of anti-inflammatory activity of drugs.

Conclusion

They increasing activity at low concentration levels but decreasing activity with high concentration. They have a critical concentration (50 mg/100ml) at which their activities are maximum. The activities of all extracts are comparable to that of Diclophenac at concentration of 50 mg/100ml. The variation of activity with time was studied at different concentration, the activities in general decreased with time.

Human keratinocyte HaCaT cells (Kim *et al.*, 2009).

Plant used:- *Acorus calamus* L.

Family:- Zingiberaceae.

Part used :- Leaves.

Common name:- Sweet flag.

Solvent:- Water.

Yield:- Not mentioned.

Dose:- 50 mg/kg.

Route of administration:- Intraperitoneally.

Chemical used:- Immunofluorescence staining, polyinosinic:polycytidylic acid.

Procedure

HaCaT cells treated with polyinosinic:polycytidylic acid (polyI:C) and peptidoglycan (PGN) induced the inflammatory reactions. The anti-inflammatory activities of ACL were investigated using RT-PCR, ELISA assay, immunoblotting, and immunofluorescence staining.

Mechanism of action

A. calamus leaf extract inhibits the production of pro-inflammatory cytokines.

Conclusion

These results suggest that ACL inhibits the production of pro-inflammatory cytokines through multiple mechanisms and may be a novel and effective anti-inflammatory agent for the treatment of skin diseases.

Leucocyte migration (Ching *et al.*, 2009).

Plant used:- *Stereospermum kunthianum*.

Family:- Bignoniaceae.

Part used:- Fresh stem bark.

Common name:- Sansami, umana and alakiriti.

Solvent:- Water.

Yield:- 26.4 % w/w.

Dose:- 500 mg/kg.

Route of administration:- Intraperitoneally.

Chemical used:- Indomethacin, Phosphate buffered saline.

Procedure

Rats were randomly allotted to groups of five animals per group. They were administered by oral route, distilled water (5 ml/kg), extract (400 mg/kg) or indomethacin (10 mg/kg) one hour before intraperitoneal injection of 0.1 ml of carrageenan, 1% w/v in normal saline. Three hours later the rats were sacrificed by excess chloroform inhalation. An incision was made on the peritoneal cavity-- of each rat and 5 ml of phosphate buffered saline was injected. After gentle massage of the peritoneum, the exudates was aspirated with a sterile syringe. The exudates volume as well as the total leucocyte count was determined.

Mechanism of action

The aqueous extract of *Stereospermum kunthianum* stem bark possesses anti-inflammatory activity which is probably related to the inhibition of prostaglandin synthesis.

Conclusion

Migration of leucocyte would not be directly related to cyclooxygenase products, but the process is inhibited by non steroidal anti-inflammatory compounds indicating that many mechanisms may be implicated in its control. The inhibitory effects of the extract on the intraperitoneal formation of exudates and leucocytes mobilization are probably due to the inhibition of prostaglandins. This possibility is reinforced by the fact that the extract at the same dose (400 mg/kg) remarkably inhibited paw oedematous process which is believed to be mediated by prostaglandine.

LPS-Activated Raw264.7 Cells (Juhas *et al.*, 2009).

Plant used:- Moutan cortex.

Family:- Paeoniaceae.

Part used:- Root cortex.

Common name:- Mudanpi.

Solvent:- Methanol.

Yield:- Not mentioned.

Dose:- 0.1 and 0.3 mg/ml.

Route of administration:- Orally.

Chemical used:- not mentioned.

Procedure

We treated LPS to Raw264.7 cells, collected media every 3 h for 24 h, detected cytokines from the collected media, and then determined appropriate time for each cytokine assay. TNF- α , IL-1 β and IL-6 were highly induced by LPS at 12, 12 and 6 h, respectively (data are not shown). TNF- α , IL-1 β and IL-6 from the culture media were analyzed at the appropriate time. 0.3 mg ml⁻¹ of MCE significantly reduced the level of TNF- α , IL-1 β and IL-6 at 12, 12 and 6 h incubation. LPS-activated PGE₂ also decreased by MCE as compared to control. PGE₂ per 1 ml medium was 6.89 pg in control and increased to 48.6 pg in LPS group. However, PGE₂ was reduced to 30.2 and 14.5 pg ml⁻¹ in 0.1 and 0.3 mg ml⁻¹ of MCE, respectively.

Mechanism of action

The anti-inflammatory activity of methanolic extract of *M. cortex* (MCE) inhibited the production of NO and prostaglandin E₂ (PGE₂), the expression of inducible NO synthase (iNOS), cyclooxygenase-2 (COX-2) and phosphorylated inhibitor of kBa (p-IkBa), and the activation of nuclear factor kB (NFkB). MCE also reduced the concentration of tumor necrosis factor- α (TNF- α), interleukin-1 β (IL-1 β) and interleukin-6 (IL-6) in the Raw264.7 cells that were activated by LPS.

Mechanism of action

The anti-inflammatory activity of aqueous extract of *Rheedia longifolia* may be due to the inhibition of release of chemical inflammatory mediators such as prostaglandins and cytokines.

Conclusion

M. cortex extract can have anti-inflammatory activity by suppressing the phosphorylation of I-kBa and the activation of NF-kB, and by inhibiting the expression of iNOS and COX-2 in LPS-activated Raw264.7 cells.

Mouse model of acute and chronic inflammation (Lee *et al.*, 2009).

Plant used:- *Asparagus cochinchinensis*.

Family:- Liliaceae.

Part used:- Dried and pulverized roots.

Common name:- Asparagus root, shiny asparagus.

Solvent:- 70% ethanol.

Yield:- Not mentioned.

Dose:- 2 g/kg.

Route of administration:- Orally.

Chemical used: Acetone, Indomethacin.

Procedure for acute model

The mouse model of acute inflammation employed here was a slight modification of a previously described *Procedure*. Edema was induced on the right ear by topical application of 2.5 μ g/ear TPA (in 20% acetone). To examine the effect of ACE on ear edema following intraperitoneal (i.p.) challenge, groups of mice were injected with ACE, vehicle (saline, negative control), or indomethacin (5 mg/kg, positive control) at 30 min prior to TPA challenge. Ear thicknesses and weights were measured 6 h after topical application of TPA.

Procedure for chronic model

The effect of ACE on chronic skin inflammation was evaluated by a slight modification of a previous described *Procedure*. Briefly, 20% of a solution of TPA (2.5 μ g/ear \times 6 times) or acetone (vehicle) were topically applied to the inner and outer ear surfaces of both ears of each mouse with a micropipette on alternate days. ACE (200mg/kg), vehicle (saline, negative control), or indomethacin (5 mg/kg, positive control) was given once a day (i.p.) for 10 days each morning immediately after TPA application. On day 10, the mice were sacrificed at 6 h after treatment, and 6-mm² diameter ear punch biopsies were collected and weighed.

Mechanism of action

The anti-inflammatory activity of *A. cochinchinensis* with 70% ethanol extract may be due to blockade of pro-inflammatory cytokine production and neutrophil-mediated myeloperoxidase activity, inhibition of IL-1 β and TNF- α and to the subsequent blockade of leukocyte accumulation.

Conclusion

These results demonstrate that ACE is an effective anti-inflammatory agent in murine phorbol ester-induced dermatitis, and suggest that the compound may have therapeutic potential in a variety of immune-related cutaneous diseases.

Neutrophil migration into peritoneal cavity

(Venegas-flores., 2002).

Plant used:- *Calea zacatechichi*.

Family:- Compositae/Asteraceae.

Part used:- Leaves.

Common name:- Dream herb, Calea.

Solvent:- Water.

Yield:- Not mentioned.

Dose:- 100 and 10 mg/kg.

Route of administration:- Intraperitoneally.

Chemical used:- Indomethacin or dexamethasone.

Procedure

The rats groups were injected with 3 ml of carrageenan (100 µg/ml) prepared in sterile saline solution into the peritoneal cavity, and 4 h later the abdominal cavity was washed with phosphate buffered saline containing 5 U/ml of heparin (Sigma, St Louis, MO) and 5% of bovine serum albumin. The total cell counts were done in a Neubauer chamber and differential cell counts were performed by the Souza and Ferreira technique. One hour before the carrageenan injection, the rats groups were treated with 10 and 100 mg/kg po of the aqueous extract, saline (control), indomethacin or dexamethasone(10 and 1 mg/kg, po), respectively.

Mechanism of action

The Anti inflammatory activity of aqueous extract of *Calea zacatechichi* may be due to inhibition of prostaglandin synthesis.

Conclusion

Dose of dexamethasone used produced a higher inhibition of neutrophil migration than the effect produced by indomethacin, supporting the possible role of leukotrienes in the inflammation model. These results suggested that antiinflammatory activity showed by *C. zacatechichi* could be related to the biosynthesis of prostaglandins and lipooxygenase products.

Paw edema induced by histamine and serotonin (Dimo *et al.*, 2006).

Plant used:- *Kalanchoe crenata*.

Family:- Crassulaceae.

Part used:- leaves.

Common name:- Never die or dog's liver.

Solvent:- Methylene chloride/ methanol.

Yield:- Not mentioned.

Dose:- 600 mg/kg.

Route of administration:- Orally.

Chemical used:- Pyrilamine malate, Histamine, Serotonin.

Procedure

In another set of experiments histamine and serotonin were used as the phlogistic agents. The n-butanol fraction of *K. crenata* extract and control vehicle were administered one hour before the injection of inflammatory mediators. The respective strength of the inflammatory mediators, The volume injected, and the time of determination of volume of edema are indicated in parentheses, serotonin and histamine. Pyrilamine maleate was used as the antagonist of histamine.

Mechanism of action

The methylene chloride/ methanol extracts of *K. crenata* and the aqueous fraction significantly inhibited paw edema induced by carrageenan in the first phase, suggesting an inhibitory effect on the release of histamine and serotonin. The n-butanol fraction showed significant inhibition of edema in all the three phases. Acute inflammation induced by formaline which provokes in production of endogenous mediators such as histamine, serotonin, prostaglandins and bradykinin.

Conclusion

K. crenata is an antiinflammatory and antiarthritic agent that blocks histamine and serotonin pathways.

Paw oedema induced by egg albumin in rat (Gertsch *et al.*, 2008).

Plant used:- *Securidaca longipedunculata*.

Family:- Poltgalacea.

Part used:- Fresh roots.

Solvent:- Petroleum ether and methanol.

Common name:- Uwar Maganinunar in Hausa and ezeogwu in Ibo.

Yield:- Methanol extract-10.9%, pet. ether fraction-0.174% and methanol fraction-12.6%.

Dose:- 50 mg/ear.

Route of administration:- Applied topically.

Chemical used:- Not mentioned.

Procedure

The methanol extract and methanol fraction Suppressed the development of paw edema by egg albumin in rats. The methanol extract evoked a non-dose related inhibition while the methanol fraction caused the reverse from 2 h. The petroleum ether fraction was did not exhibit anti-inflmmatory activity.

Mechanism of action

Pharmacological screening of root bark extracts of *S. longipedunculata* has revealed that the root bark possesses potent anti-inflammatory effect in the topical and systemic models of acute inflammation. These extracts may have inhibited the release

of pro-inflammatory mediators of acute inflammation such as histamine and prostaglandin.

Conclusion

These extracts may have inhibited the release of pro-inflammatory mediators of acute inflammation such as histamine and prostaglandin.

PGE₂ induced hind paw edema (Kupeli *et al.*, 2007).

Plant used:- *Geranium pretense subsp. Finitimum*.

Family:- Geraniaceae.

Part used:- Aerial parts.

Common name:- Woronouu.

Solvent:- Water.

Yield:- Not mentioned.

Dose:- 0.5 mg/25µl.

Route of administration:- Intraperitoneally.

Chemical used:- ProstaglandinE₂ in Tyroid's solution.

Procedure

Sixty minutes after the oral administration of a test sample or dosing vehicle, each mouse was injected with freshly prepared suspension of PGE₂ in Tyroid's solution into subplantar tissue of the right hind paw. As the control, 5µl Tyroid's solution was injected into that of the left hind paw. Paw edema was measured in every 15 min during a period of 75 min after the induction of inflammation.

Mechanism of action

The anti-inflammatory activity of aqueous extract of *Geranium pretense subsp. Finitimum* may be due to the inhibition of production of prostaglandin. It also reduces the TPA induced mouse ear edema.

Conclusion

In this study revealed that the aerial parts of *Geranium pratense subsp. finitimum* possess anti-inflammatory and antinociceptive properties and contribute a further support its use in traditional medicine for alleviating painful inflammatory conditions.

Pleurisy induced by LPS in mice (Frutuoso *et al.*, 2007).

Plant used:- *Rheedia longifolia*.

Family:- Clusiaceae.

Part used:- Leaves.

Common name:- Bacupari.

Solvent:- Water.

Yield:- Not mentioned.

Dose:- 50 mg/kg.

Route of administration:- Intraperitoneally.

Chemical used:- Acetic acid, cytosmears stained, May-Grunwald-Giemsa dye.

Procedure

The leaves aqueous extract of *R. longifolia* (50 mg/kg, i.p.) was administered 1 h before the injection of LPS. Six hours after the injection of LPS the animals were sacrificed in a CO₂ chamber, the thoracic cavity was opened and washed with 1 ml of PBS containing heparin 20 UI. The pleural wash was collected with the aid of a pipette and used to analyze the cellular infiltrate. The total number of leukocytes collected in pleural wash was evaluated in a Neubauer chamber after dilution of a pleural wash sample in 2% acetic acid. The differential analysis of leukocytes was accomplished in cytosmears stained with May-Grunwald-Giemsa dye. Each experimental group was performed with at least eight animals.

Mechanism of action

The anti-inflammatory activity of aqueous extract of *R. longifolia* may be due to the inhibition of release of chemical inflammatory mediators such as prostaglandins and cytokines.

Conclusion

R. longifolia LAE inhibited an inflammatory reaction induced by LPS in the pleural cavity of mice. Acute toxicity was evaluated in mice treated with the extract for seven days with 50 mg/kg/day. Neither death, nor alterations in weight, blood leukocyte counts or hematocrit were noted. Our results suggest that aqueous extract from *R. longifolia* leaves has analgesic and anti-inflammatory activity with minimal toxicity.

Topical acute edema of the mouse ear (Gertsch *et al.*, 2008).

Plant used:- *Securidaca longipedunculata*.

Family:- Poltgalacea.

Part used:- Fresh roots.

Common name:- Uwar Maganinunar in Hausa and ezeogwu in Ibo.

Solvent:- Petroleum ether and methanol.

Yield:- Methanol extract-10.9%, pet. ether fraction-0.174% and methanol fraction-12.6%.

Dose:- 50 mg/ear.

Route of administration:- Applied topically.

Chemical used:- Not mentioned.

Procedure

The extract and fractions inhibited topical edema induced by xylem in the mouse ear. The fraction caused greater than the methanol extract in the order Petroleum ether fraction > Methanol fraction > Methanol extract.

Mechanism of action

Pharmacological screening of root bark extracts of *S. longipedunculata* has revealed that it possesses potent anti-inflammatory effect in the topical and systemic models of acute inflammation. These extracts may have inhibited the release of pro-inflammatory mediators of acute inflammation such as histamine and prostaglandin.

Conclusion

Pharmacological screening of root bark extracts of *S. longipedunculata* has revealed that the root bark possesses potent anti-inflammatory effect in the topical and systemic models of acute inflammation. These extracts may have inhibited the release of pro-inflammatory mediators of acute inflammation such as histamine and prostaglandin.

TPA-treated mouse ear edema (Hargrove *et al.*, 2008).

Plant used:- *Polygonum cuspidatum*.

Family:- Polygonaceae.

Part used:- Root.

Common name:- Japanese knotweed or Mexican bamboo.

Solvent:- 50 % ethanol.

Yield:- Not mentioned.

Dose:- 2.5 mg/ear.

Route of administration:- Topically.

Chemical used:- 50% ethanol, Indomethacin.

Procedure

Edema was induced in both ears of each mouse by the topical application of 2 µg TPA dissolved in 20 µL of acetone to both the inner and outer ear surfaces. Thirty minutes after the application of TPA, the inner and outer surface of each ear was treated (10 µL to each side) with 50% ethanolic solutions of PCE in doses of 0.075, 0.15, 0.3, 1.25 and 2.5 mg PCE/ear (n = 8 at each dosage). Comparisons included equal volumes of 50% ethanol (vehicle control), indomethacin (0.5 mg/ear dissolved in 50% ethanol as an anti-inflammatory drug standard), or a 50% ethanol solution of trans-3, 5, 4'-trihydroxystilbene (resveratrol, 0.6 mg/ear). The thickness of each ear was measured using a micrometer before and at 4 h and 24 h after TPA administration. The micrometer was applied near the top of the ear distal to the cartilaginous ridges. At 24 h each animal was sacrificed with CO₂ inhalation by the IACUC approved protocol. Ear punch biopsies (6 mm diameter hole punch) were taken immediately, weighed, frozen and stored at -80°C. A single investigator performed all ear measurements and biopsies in order to standardize the *Procedure* and reduce experimental error.

Mechanism of action

P. cuspidatum extract inhibits development of edema and neutrophil infiltration in the TPA-treated mouse ear model of topical inflammation.

Conclusion

PCE inhibits development of edema and neutrophil infiltration in the TPA- treated mouse ear model of topical inflammation.

Xylen or croton oil-induced mouse ear edema (Yin *et al.*, 2000).

Plant used:- *Boschnikia rossica*.

Family:- Betulaceae.

Part used:- Whole plant.

Common name:- Bu Lao Cao.

Solvent:- Methanol extract was fractioned with CH₂Cl₂ and H₂O.

Yield:- Not mentioned.

Dose:- 500-1000 mg/kg.

Route of administration:- Orally.

Chemical used:- BRH₂O extract.

Procedure

An edema was induced on the right ear by topical application of xylen in mice 30 minutes after oral administration of 500mg/kg-1000mg/kg BR-H₂O extract or BR-CH₂Cl₂ extract. The left ear was controlled. Ear edema was measured by comparing the difference in weight (mg) between the same size of left and right ears 30 minutes after xylen induction and 4h after croton oil-induction of inflammation and swelling degree and inhibition rate were calculated.

Mechanism of action

B. rossica extract exhibited inhibitory effect on formation of preneoplastic hepatic foci in early stage of rat chemical hepatocarcinogenesis. Both CH₂Cl₂ and H₂O extracts from *B. rossica* exerted anti-inflammatory effect in rats and mice.

Conclusion

B. rossica extract exhibited inhibitory effect on formation of preneoplastic hepatic foci in early stage of rat chemical hepatocarcinogenesis. Both CH₂Cl₂ and H₂O extracts from *B. rossica* exerted anti-inflammatory effect in rats and mice.

Xylene induced ear edema (Hosseinzadeh *et al.*, 2002).

Plant used:- *Crocus sativus* Linn.

Family:- Iridaceae.

Part used:- Powder of stigma and petal.

Common name:- Saffron.

Solvent:- Water and ethanol.

Yield:- For stigma, water-50.8%w/w and ethanol-56.6%w/w and for petal, water 15.5%w/w and ethanol-19%w/w.

Dose:- 0.8 g/kg and 2 g/kg.

Route of administration:- Intraperitoneally.

Chemical used:- Diclofenac, Dexamethasone.

Procedure

Mice were divided into groups of seven. Thirty minutes after Intraperitoneally injection of the extract, diclofenac and dexamethasone, 0.03 ml of xylene was applied to the anterior and posterior surfaces of the right ear. The left ear was considered as control. Two hours after xylene application. mice were killed and both ears were removed. Circular sections were taken, using a cork borer with a diameter of 7 min, and weighed. The increased in weight caused by the irritant was measured by subtracting the weight of the untreated left ear section from that of the treated right ear sections.

Mechanism of action

The stigma extracts showed activity against acute and chronic inflammation. The petal extracts showed anti-inflammatory activity in the chronic inflammation. The anti-inflammatory activity of the petal extracts may be due to their content of flavonoids, tannins and anthocyanins which inhibits the cyclooxygenase activity.

Conclusion

The aqueous and ethanolic extracts of saffron stigma and petal have an antinociceptive effect, as well as acute and/or chronic anti-inflammatory activity.

Zymosan-induced oedema in mice (Stefanova *et al.*, 1995).

Plant used:- *Fraxinus ornus* Linn.

Family:- Oleaceae.

Part used:- Stem bark.

Common name:- Flowering Ash, Manna, Manna Ash.

Solvent:- 96% boiling ethanol.

Yield:- Not mentioned.

Dose:- 5-15 mg/kg.

Route of administration:- Intraperitoneally.

Chemical used:- 2% Zymosan.

Procedure

Oedema in ICR mice was induced by injecting 0.025 ml of 2% Zymosan suspension in saline into the left hind paw. The right hind paw was used as a control and was injected with 0.025 ml of 0.9% saline. The animal were killed 3 h later, both hind paw cut off at the ankle and the difference between their weights calculated.

Mechanism of action

F. ornus bark contains several hydroxycoumarin derivatives, the main one being esculin (EN). The total ethanol extract from *F. ornus* bark contains substances of high potency capable of inhibiting classical pathway and alternative pathway complement activity. The comparison between the effects obtained with total ethanol extract and esculin in the haemolytic inhibitory assay indicates that the anticomplementary action of TE is not due only to EN.

Conclusion

Zymosan- induced AP activation of mouse serum in vitro is strongly suppressed by TE and EN. The effective dose of about 5-15 mg/kg is comparable to the data known for other lipooxygenase inhibitors like phenidone.

 λ - Carrageenan (CARR) - induced paw edema (Huang *et al.*, 2008).

Plant used:- *Hydrocotyle batrachium*s.

Family:- Apiaceae.

Part used:- Whole plant.

Common name:- Marshpennywort.

Solvent:- Water.

Yield:- 26.37% w/w.

Dose:- 500 and 100 mg/kg.

Route of administration:- Orally.

Chemical used:- Indomethacin.

Procedure

The anti-inflammatory activity of HBW (*Hydrocotyle batrachium* Hance) was determined by the λ - Carrageenan-(CARR) induced edema test. Male ICR mice were randomly assigned to five groups and then fasted with free access to water for 24 hours before the experiment. Fifty microliters of a 1% suspension of CARR in saline, which had been prepared 30 min before each experiment, was injected into the plantar side of the right hind paw of the mice. After 60 min, HBW at dose of 100, 500 and 1000 mg/kg were administered orally, and after 90 min, indomethacin at the dose of 10 mg/kg was administered via an intraperitoneal route after the CARR treatment. Paw volume was measured prior to CARR injection and at 60, 120, 180, 240, 300, 360 min interval after the administration of the endematogenic agent using a plethysmometer. The degree of swelling was evaluated according to the a-b value, where a is the volume of the right hind paw after CARR treatment and b is the volume of the right hind paw before CARR treatment. Indomethacin was used as a positive control compound.

Mechanism of action

The anti-inflammatory activity of water extract of *H. batrachium* may be due to the inhibition of mediators of inflammation and also inhibits the synthesis of prostaglandin.

Conclusion

HBW appears to have analgesic and anti-inflammatory activities.

Carrageenan and dextran-induced pedal edema (Sarkar *et al.*, 2005).

Plant used:- *Aloe vera*.

Family:- Asphodelaceae.

Part used:- Leaves.

Common name:- Kumari, Musabbar.

Solvent:- Dimethylsulfoxide.

Yield:- Not mentioned.

Dose:- Upto 25mg/kg.

Route of administration:- Orally.

Chemical used:- Dextran, Indomethacin.

Procedure

Animals were divided into four groups (n = 6). In all the groups, acute inflammation was induced by injection of either 0.1 ml of freshly prepared carrageenan (1.0 %) in 0.9 % w/v NaCl solution or 0.1 ml of dextran (2%) in 0.9% w/v NaCl solution into the subplantar region of the hind paw of rats. AVL in different doses (0- 25 mg/kg) and indomethacin (2.5, 5 and 10 mg/kg, as a standard reference) were orally administered one hour before injection of the phlogistic agent. The paw volumes were measured plethysmographically (using water) once prior to administration of the phlogistic agent and thereafter at hourly intervals for 3 h.

Mechanism of action

The anti-inflammatory activity that is mediated through antibradikinin activity and inhibition of prostaglandin production. *A. vera* leaf exudate also demonstrated that this anti-inflammatory activity is mediated partly via reduction of nitric oxide production in macrophages.

Conclusion

A. vera possesses acute and chronic antiinflammatory activity, which is partly mediated by reduced production of NO, which in turn prevents the release of inflammatory mediators.

Freund's adjuvant-induced model (Sarkar *et al.*, 2005).

Plant used:- *Aloe vera*.

Family:- Asphodelaceae.

Part used:- Leaves.

Common name:- Kumari, Musabbar.

Solvent:- Dimethylsulfoxide.

Yield:- Not mentioned.

Dose:- Upto 25mg/kg.

Route of administration:- Orally.

Chemical used:- Dextran, Indomethacin.

Procedure

Rats were divided into three groups (n = 6). Briefly, 0.1 ml of Freund's complete adjuvant was injected intradermally into the plantar aspect of the hind paw of each animal. Animals were administered AVL (25 mg/kg, orally) and dexamethasone (0.1 mg/kg, orally, as a standard reference) for the initial 13 days. The degree of inflammation was measured plethysmographically; accordingly, edema formation and the percentage of inhibition was calculated as described above on days 1, 3, 5, 9, 13 and 21 and the primary and the secondary lesions were measured. Primary lesions refer to the edema formation in the injected hind paw that peaks 3-5 days after injection of the phlogistic agent and is measured on day 5. Secondary lesions are immunologically mediated changes characterized by inflammation of the non-injected sites (hindleg, forepaw, ears nose and tail) decrease in weight and occur after a delay of 11-12 days. Accordingly, secondary lesions were evaluated by calculating the percent inhibition of paw volume of the non-injected right paw over control on day 21 and using an arthritic index as the sum of scores according to the method of Schorlemmer.

Mechanism of action

The anti-inflammatory activity that is mediated through antibradikinin activity and inhibition of prostaglandin production. *Aloe vera* leaf exudates also demonstrated that this anti-inflammatory activity is mediated partly via reduction of nitric oxide production in macrophages.

Conclusion

A. vera possesses acute and chronic antiinflammatory activity, which is partly mediated by reduced production of NO, which in turn prevents the release of inflammatory mediators.

Kaolin- induced paw oedema (Sevastre *et al.*, 2007).

Plant used:- *Peucedanum officinale*.

Family:- Apiaceae.

Part used:- Whole plant.

Common name:- Hog's fennel.

Solvent:- Not mentioned.

Yield:- Not mentioned.

Dose:- 25 mg/kg.

Route of administration:- Orally.

Chemical used:- kaolin injection.

Procedure

One hour later, suspension of kaolin 10% was intraplantary injected. The measurements, made by UGO BASILE 7140 digital plethysmometer, were done before kaolin injection (for assessing the initial volume of paw), and in 2, 4 and 24 hours after the injection. Inflammatory oedema represent the difference between the injected paw volume at 2, 4, respectively 24 hours and initial volume (iV). The inhibition of oedema (IO) represents the difference in paw volume, between treated animals (positive control group and experimental group) and untreated ones (negative control).

Mechanism of action

Peucedanum species contains mainly coumarins. Coumarins highly effective in the treatment of inflammatory oedema. It also acts as inhibitors of cyclooxygenase and 5-lipoxygenase. The *Peucedanum* extract have agonistic effect of histamine and down regulation of cyclooxygenase activity. It also inhibits the release of prostaglandin synthesis.

Conclusion

P. officinale has proved a significant antiinflammatory activity on kaolin induced rat paw oedema model, however the efficiency has been found lower than classic antiinflammatory therapy (phenylbutazone).

Lipopolysaccharide induced septic shock (Rao *et al.*, 2007).

Plant used:- *Rhododendron dauricum* Linn.

Family:- Ericaceae.

Part used:- Leaves.

Common name:- Ezomurasaki shakunage.

Solvent:- Water.

Yield:- 16.2% w/w.

Dose:- 200-500 mg/kg.

Route of administration:- Orally.

Chemical used:- D-galactosamine, *Rhododendron dauricum* L. extracts, alanine, aspartate aminotransferase.

Procedure

In this, Number of deaths per group, Serum level of alanine aminotransferase, Serum level of aspartate aminotransferase. No deaths were recorded in the group of mice administered with only saline and D-galactosamine. However, in galactosamine primed mice injected with Lipopolysaccharide

(LPS), all the animals died before the completion of the experiment. The number of deaths in the group of animals pretreated with 10, 20, 40, and 80 mg/kg of the *Rhododendron dauricum* L. extracts prior to LPS-challenge were three, two, one and none, respectively. No deaths were recorded in the group of mice that received pentoxifylline (100 mg/kg). D-galactosamine injection before saline treatment resulted in serum level of 270.7 ± 3.2 U/ml and 304.8 ± 6.6 U/ml of alanine and aspartate aminotransferase respectively. In D-galactosamine primed mice injected with LPS, the levels elevated to 297.8 ± 3.5 U/ml and 335.8 ± 7.8 U/ml for alanine and aspartate aminotransferase respectively.

Mechanism of action

The exact *Mechanism of action* is not known.

Conclusion

The effect of *R. dauricum* L. extract was tested on induced dye leakage in the mouse skin. The effect of extract and pentoxifylline were evaluated 2 hr after injection of LPS. The extract was found to produced inhibition of dye leakage to the same extent as pentoxifylline. Subcutaneous injection of LPS on the back of mice and rats induced a plasma leakage at the site of injection and thus is used as a model of inflammation. It is also determine cytokines.

Plasma leakage in the mouse skin (Rao *et al.*, 2007).

Plant used:- *Rhododendron dauricum* Linn.

Family:- Ericaceae.

Part used :- Leaves.

Common name:- Ezomurasaki shakunage.

Solvent:- Water.

Yield:- 16.2% w/w.

Dose:- 200-500 mg/kg.

Route of administration:- Orally.

Chemical used:- *Rhododendron dauricum* L. extract.

Procedure

The effects of the extracts on LPS- induced dye leakage were evaluated 2 hr after subcutaneous injection of LPS. Pretreatment with *Rhododendron dauricum* L. extract produced a dose related inhibition of dye leakage. Dye leakage in mice given only saline before LPS injection was 57.3 ± 4.9 μ g/g wet weight. This was significantly reduced to 38.0 ± 2.8 μ g/g wet weight by the extract given at 10 mg/kg.

Mechanism of action

The exact *Mechanism of action* is not known.

Conclusion

The effect of *R. dauricum* L. extract was tested on induced dye leakage in the mouse skin. The effect of extract and pentoxifylline were evaluated 2 hr after injection of LPS. The extract was found to produced inhibition of dye leakage to the

same extent as pentoxifylline. Subcutaneous injection of LPS on the back of mice and rats induced a plasma leakage at the site of injection and thus is used as a model of inflammation.

Hyaluronidase inhibitory assay (Hayati *et al.*, 2007).

Plant used:- *Prismatomeris malayana*.

Family:- Rubiaceae.

Common name:- *Prismatomeris albidiflora* King.

Part used:- Root, stem and leaf.

Solvent:- solution of leaf crud.

Yield:- 9.84 % w/v.

Dose:- 200 mg/kg.

Route of administration:- Orally.

Chemical used:- Sodium chloride, Hyaluronic acid, Acetic acid,

Procedure

The assay medium consisting of 1.00 to 1.67 U hyaluronidase in 100 μ l 20 mM sodium phosphate buffer pH 7.0 with 77 mM sodium chloride and 0.01% Bovine Serum Albumin (BSA) was pre-incubated with 5 μ l of the test compound in DMSO (dimethyl sulphoxide) for 10 min at 37 °C. Then the assay was commenced by adding 100 μ l hyaluronic acid (0.03% in 300 mM sodium phosphate, pH 5.35) to the incubation mixture and incubated for a further 45 min at 37°C. The undigested hyaluronic acid was precipitated with 1 ml acid albumin solution made up of 0.1% bovine serum albumin in 24 mM sodium acetate and 79 mM acetic acid, pH 3.75. After standing at room temperature for 10 min, the absorbance of the reaction mixture was measured at 600 nm. The absorbance in the absence of enzyme was used as the reference value for maximum inhibition. The inhibitory activity of test compound was calculated as the percentage ratio of the absorbance in the presence of test compound vs. absorbance in the absence of enzyme. The enzyme activity was checked by control experiment that ran simultaneously, in which the enzyme was pre-incubated with 5 μ l DMSO, followed by the assay *Procedures* described above. In this case, the percentage ratio of the absorbance in the presence of enzyme vs. that in the absence of enzyme was in the range of 15 to 20%.

Mechanism of action

Hyaluronidase is an enzyme that degrades hyaluronic acid and chondroitin sulfate which are components of the extra cellular matrix of connective tissue. By degrading the components of connective tissue, hyaluronidase promotes the spread of inflammatory mediators throughout these tissues, thereby contributing to the pathogenesis of inflammatory diseases such as allergic effects, migration of cancer cells, inflammation and the increase in permeability of vascular system.

Conclusion

The leaves (PML) showed the highest activity on hyaluronidase inhibitory assay compared to the roots (PMR) and stem (PMS).

SUMMARY AND CONCLUSION

The detail literature survey was done on various plants which can be used in inflammation including their plant used, family, part used, common name, solvent of extraction, yield, dose, route of administration, chemical used, *Procedure Mechanism of action*, and *Conclusion* was done. It is cleared from *Mechanism of action* of these extract that almost all drugs work on the principle of inhibition of prostaglandin and other inflammatory mediators synthesis.

REFERENCES

- Amresh, G., Zeashan, H., Rao, Ch, V., Singh, P, N., Prostaglandin mediated anti-inflammatory and analgesic activity of *Cissampelos pareira*. *Acta Pharmaceutica Scientia* 2007; 49: 153-160.
- Arunachalam, G., Subramanian N., Pazhani G, P., Ravichandran V., Anti inflammatory activity of methanolic extract of *Eclipta prostrata* L. (Asteraceae). *African journal of pharmacy and pharmacology*. 2009; 3: 97-100.
- Badilla, B., Arias, Y., Mora, G. A., and Poveda, L. J., Anti inflammatory and antinociceptive activities of *Loasa speciosa* in rats and mice. *Fitoterapia*. 2003; 74: 45-51.
- Segura, L., Vila, R., Gupta, M, P., Espósito-Avella M., Adzet, T., Cañigueral, S., Anti inflammatory activity of *Anthurium cerrocampaense*. *Journal of ethnopharmacology*. 1998; 61: 243-248.
- Ching, F, P., Omogbai, E, K, I., Okpo, S, O., Ozolua, R, I., Antiinflammatory activity of aqueous extract of *Stereospermum kunthianum* (cham, Sandrine petit) stem bark in rats. *Indian journal of pharmaceutical sciences*. 2009; 71: 106-110.
- Chun, S., Jee, S., Lee, S. Anti-inflammatory activity of the methanol extract of *Moutan cortex* in LPS- activated Raw264.7 cells. *eCAM*, 2006; 1-7.
- Dimo, T., *et al.* Anti-inflammatory activity of leaf extracts of *Kalanchoe crenata* Andr. *Indian Journal of Pharmacology*. 2006; 38: 115-119.
- Frutuoso, V, S., *et al.* Analgesic and antiinflammatory activity of the aqueous extract of *Rheedia longifolia* Planch & Triana. *Memorias do instituto oswaldo cruz*. 2007; 102.
- Gertsch *et al.* Beta- caryophyllene is a dietary cannabinoid. *Proceedings of the national academy of sciences* (2008) 9099.
- Ghule, B, V., *et al.* Analgesic and anti inflammatory activities of *Lagenaria siceraria* stand. Fruit juice extract in rats and mice. *Pharmacognosy Magazine*. 2006; 2: 232-236.
- Hargrove, J, L., *et al.* Topical anti inflammatory activity of *Polygonum cuspidatum* extract in the TPA model of mouse ear inflammation. *Journal of inflammation*. 2008; 1-7.
- Hemamalini K., K. Om Prasad Naik, Ashok, P. Anti inflammatory and analgesic effect of methanolic extract of *Anogeissus acuminata* leaf. *Int J Pharm Biomed Res* 2010; 1(3): 98-101.
- Hosseinzadeh, H., Younesi, H, M., Anti nociceptive and anti inflammatory effects of *Crocus sativus* L. stigma and petal extracts in mice. *BMC pharmacology* (2002). 2.
- Huang, S., *et al.* Analgesic and anti-inflammatory activities of an aqueous extract of *Hydrocotyle batrachium* hance in mice. 2008; 179-184.
- Ilavarasan, R., Mallaika, M., Venkataraman, S., Anti-inflammatory and antioxidant activities of *Cassia fistula* Linn. bark extracts. *African Journal Traditional, complementary and alternative medicines*. 2005; 2: 70-85.
- Jain, R., *et al.* Anti- inflammatory action of *Abutilon indicum* (L.) sweet leaves by HRBC membrane stabilization. *Research Journal of Pharmacy and Technology*. 2009; 2: 415-416.
- Kim, H., Han, T, H., Lee, G, S., Anti inflammatory activity of a water extract of *Acorus calamus* L. leaves on keratinocyte HaCaT cells. *Journal of ethnopharmacology*. 2009; 122: 149-156.
- Kupeli, E., *et al.* Estimation of anti nociceptive and anti inflammatory activity on *Geranium pratense* subsp. *Finitimum* and its phenolic compounds. *Journal of ethnopharmacology*. 2007; 114: 234-240.
- Lee, D, Y., *et al.* Anti- inflammatory effects of *Asparagus cochinchinensis* extract in acute and chronic cutaneous inflammation. *Journal of Ethnopharmacology*. 2009; 121: 28-34.
- Manga, H, M., *et al.* In vivo anti inflammatory activities of *Alchornea cordifolia* (Schumach. & Thonn.) Mull. Arg. (Euphorbiaceae). *Journal of ethnopharmacology*. 2004; 92: 209-214.
- Nor Hayati, A., *et al.* Bioassay guided fractionation and isolation of anti-inflammatory compound from *Prismatomeris malayana* Ridley. 2007; 27-32.
- Paschapur, M, S., *et al.* Evaluation of anti-inflammatory activity of ethanolic extracts of *Borassus fabbellifer* L. male flowers (inflorescences) in experimental animals. *Journal of Medicinal Plants Research*. 2009; 3: 049-054.
- Rao, H, S., Singh, B., Singh, N., Singh, R., Anti inflammatory properties of *Rhododendron dauricum* Linn. Inhibition of lipopolysaccharide induced septic shock and vascular permeability. *Pharmacognosy Magazine*. 2007; 3: 232-235.
- Sarkar, D., *et al.* Effect of Aloe vera on nitric oxide production by macrophages during inflammation. *Indian journal of pharmacology*. 2005; 37: 371-375.
- Sevastre, B., *et al.* Anti inflammatory activity of *Peucedanum officinale* on rats. *Bulletin USAMV-CN*. 2007; 64: 295-298.
- Stefanova, Z., *et al.* Effect of a total extract from *Fraxinus ornus* stem bark and esculin on zymosan- and carrageenan-induced paw oedema in mice. *Journal of ethnopharmacology* 1995; 46: 101-106.
- Venegas-flores, H., Segura-cobos, D., Vazquez-cruz, B., Anti inflammatory activity of aqueous extract of *Calea zacatechichi*. *Proc. West Pharmacol. Soc*. 2002; 45: 110-111.
- Yin, Z, Z., *et al.* Effect of *Boschniakia rossica* on expression of GSTP, p53 and p21ras proteins in early stage of chemical hepatocarcinogenesis and its anti-inflammatory activities in rats. *World Journal of Gastroentero*. 2000; 6: 812-818.