Research Article

In Vitro Anti Inflammatory and Anti Arthritic Activity of Selected Medicinal Plant

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ABSTRACT

The aim of the study is to carry out the in vitro antiarthritic and anti-inflammatory activity in aerial parts of *Myxopyrum Serratulum* A.W Hill. *In vitro* antiarthritic activity was carried out using protein Denaturation Method and *In vitro* Anti Inflammatory Activity was Carried Out Using HRBC Membrane Stabilization Method. The percentage stabilization of ethanolic extracts on HRBC Membrane stabilization was found 61.25% at 200 µg/ml. The percentage inhibition of ethanolic extracts on Protein Denaturation method was found to be 67.51% at 200 µg/ml. *In vitro* studies which was carried out by the above mentioned methods brings out the fact that the ethanolic extract possess better activity which was comparable to the standard drug (DICLOFENAC SODIUM).

Keywords: anti-inflammatory activity, diclofenac sodium, medicinal plant.

INTRODUCTION

Herbal medicine is unwritten science is well established in some cultures and tradition of developing countries. Arthritic conditions are treated using traditional medicine with considerable success. Although various modern drugs are used to treat these type of disorders their prolonged usage may cause severe side effects .So there is an urge to develop new therapeutic agents with minimum side effects. *Myxopyrum serratulum* A.W Hill (Oleaceae) otherwise called caturamullai in Tamil is widely distributed throughout Kerala in evergreen forest at altitudes of 600-900m.It is widely used in the treatment of cuts, wounds, scabies, cough, nerve complaints, head ache, back ache, arthritis[1]. A Perusal of literature revealed that the leaves of *Myxopyrum serratulum* A.W Hill have been reported for preliminary phytochemical screening indicated the presence of flavonoids, terpenoids, glycosides, tannin, saponin[2] etc, GC-MS analysis has been carried out[3], *In vitro* antioxidant activity has been performed[4], quantification of phytoconstituents was carried out[5], further isolation of iridoid glycosides has been carried out.[7] Since, so far no scientific evaluation has been made to carry out *in vitro* anti-inflammatory and antiarthrhitic activity in *Myxopyrum serratulum* A .W Hill (Aerial, parts). The current study is focused to determine the *in vitro* activities in selected medicinal plant.

MATERIALS AND METHODS

The fresh aerial parts were collected from Tirunelvelli in month of September and it was authenticated by Dr.V.Chelladurai, CCRAS, Tirunelvelli. The voucher specimen is deposited in Department of Pharmaceutical chemistry, Faculty of Pharmacy for future reference. The collected aerial parts were shade dried and coarsely powdered. The Plant material were subjected to successive extraction using petroleum ether, chloroform, ethyl acetate, ethanol as solvents by cold maceration process. The solvents were distilled under reduced pressure using rotary evaporator.

In vitro antiarthritic activity

Protein Denaturation Method [8]

1. Test solution (0.5ml): It consist of 0.05ml of test solution of various concentrations (10-1000 µg/ml) and 0.45ml of Bovine serum albumin (5% aqueous solution)

2. Test control solution (0.5ml): It consist of 0.05ml of distilled water and 0.45ml of Bovine serum albumin (5% aqueous solution).

3. Product control (0.5ml): It consist of 0.05ml test solution of various concentrations (10-1000µg/ml) and 0.45ml of distilled water.

4. Standard solution (0.5ml): It consist of 0.05ml of Diclofenac sodium (100, 250, 1000µg/ml) and 0.45ml of Bovine serum albumin (5%aqueous solution).

PH was adjusted to 6.3 to all above solution by using 1N HCl. All the sample solution was incubated at 37^{O}C for 20 minutes and the temperature was increased to 57 ^{o}C for 3 minutes. Allow the solution to cool for some time then add 2.5ml of Phosphate buffer to all above solution. The absorbance of the resulting solution is measured at 416 nm using UV visible spectrophotometer. The Percentage inhibition of protein denaturation was calculated as per the given formula.

$$\textit{Percentage Inhibition} = 100 - \frac{(\text{O D of test solution} - \text{O D of Product control})}{\text{O D of test control}} \times 100$$

In vitro anti-inflammatory activity

HRBC Membrane Stabilisation Method [9]

The Human red blood cell membrane stabilisation was used to determine the *in vitro* anti-inflammatory activity.

The blood sample was collected from healthy human volunteers (who has not consumed any NSAIDS for period of 2 weeks).Collected blood sample was mixed with equal volume of Alsever solution(2% dextrose, 0.8% sodium citrate, 0.5% Citric acid and 0.42% of Sodium chloride) and the centrifuge at 3000 rpm. The packed cells were washed with isosaline and 10% suspension was made. Different concentration of the extracts were prepared (10-1000µg/ml) using DMSO to all the above solution add 1ml Phosphate buffer, 2ml of hyposaline and 0.5ml of HRBC Suspension. All the above solution were incubated 37 ^{o}C for 30 minutes and centrifuge at 3000 rpm for 20 minutes. The clear supernatant liquid was estimated using UV visible spectrophtometer at 560 nm. Reference standard used are Diclofenac sodium

RESULTS AND DISCUSSION

Inflammation is a common phenomenon and it is a reaction of living issue towards injury. Phytochemical screening indicates the presence of flavonoids, tannin, saponins, glycosides etc. In this current research work *in vitro* anti-inflammatory, antiarthritic activity was performed using HRBC Membrane stabilization method and protein Denaturation method. HRBC is similar to lysosomal membrane and its stabilization implies that the extract may as well stabilize lysosomal membrane.

Table 1: Effect of *Myxopyrum serratulum* A.W hill on HRBC membrane stabilization method and protein denaturation method

Concentration (200µg/ml)	% stabilization	% inhibition
Petroleum ether extract	21.32	25.38
Chloroform extract	35.46	33.46
Ethyl acetate extract	48.24	47.25
Ethanol extract	61.25	67.51
Diclofenac sodium	70.52	72.64

In vitro anti-inflammatory activity

HRBC membrane stabilization method

The *in vitro* anti-inflammatory method involves the stabilization involves stabilization of HRBC Membrane by hypotonicity induced membrane lysis. The percentage protection of ethanolic extracts was found 61.25% at 200 µg/ml.

In vitro antiarthiritc activity

Protein Denaturation method

The production of auto antigen in certain arthritic disease may be due to denaturation of protein. Percentage inhibition of ethanolic extract was found to be 67.51% at 200 µg/ml and the effect was compared with standard drug (Diclofenac sodium). The results emphasize that the ethanolic extract was capable of controlling the production of autoantigen and inhibits denaturation of protein

CONCLUSION

In-vitro studies which were carried out by the above mentioned methods proved that the ethanolic extracts possess anti-inflammatory and antiarthritic activity which was similar to that of standard.

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REFERENCES

1. Vaidhyrathnam Ps Varriers, Indian medicinal plants compendium of 500 species, Arya vaidya sala, Kottakkal, 4, 1996, 882.
2. Gopalakrishnan S, Rajameena R and Vadivel E, Phytochemical and Pharmacognostic studies of leaves of Myxopyrum serrtulum A.W Hill, Journal of chemical and pharmaceutical research, 4(1), 2012, 788-794.
3. Gopalakrishnan S, A Rajameena. R and Vadivel. E, antimicrobial activity of leaves of Myxopyrum serratulum A.W. Hill, International journal of Pharmaceutical sciences and drug research, 4(1), 2012, 31-3.
4. Sheelarani T, Gopal V, Chitra K, Seethalakshmi S, Invitro antioxidant activity of Myxopyrum serratulum A.W HILL, international journal of pharmacy and pharmaceutical sciences, 5(4), 2013, 545-546
5. Sheelarani T, Gopal V, Chitra K. Seethalakshmi, Quantification and screening of Phytoconstituents in selected medicinal plant, Research journal of pharmaceutical technology, 6(11), Nov 2013, 1235.
6. Franzyk, Iridoid glucosides from myxopyrum smilacifolium, *Journal of Natural products,* 64(5), 2001, 632-633.
7. Chanda S. Dave R, Kaneria M and Nagani K, Invitro models for antioxidant activity evaluation of some medicinal plants possessing antioxidant properties, *African journal of microbiology Research,* Dec 2009 3(13), 981-996.
8. Ravi Rajurkar, Ritesh jain, narendra matake Prshant Aswar, SS. Khadabi., Anti-inflammatory action of Abuliton indicum Sweet leaves by HRBC Membrane Stabilization, *Research journal of pharmacy and Technology*, 2 (2), June 2009, 415-416.

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