doi.org/10.36721/PJPS.2022.35.4.SP.1281-1286.1

Evaluation of anti-inflammatory and analgesic effects of *Hertia intermedia* (Boiss.) Kuntze extract

Mohammad Younis[1], Javeid Iqbal[1], Shafi Muhammad[2], Syed Umer Jan[3], Abdul Jabbar[2], Mehjabeen[4], Abdul Qadir[3], Nagina Soomer Khan[2], Muhammad Arslan[2], Pervaiz Akhtar Shah[5] and Zahid Khan[6]

[1]Department of Pharmacology, Faculty of Pharmacy and Health Sciences, University of Balochistan, Quetta, Pakistan
[2]Department of Pharmacognosy, Faculty of Pharmacy and Health Sciences, University of Balochistan, Quetta, Pakistan
[3]Department of Pharmaceutics, Faculty of Pharmacy and Health Sciences, University of Balochistan, Quetta, Pakistan
[4]Department of Pharmacology, Faculty of Pharmacy, Federal Urdu University of Arts, Science and Technology, Karachi, Pakistan
[5]Punjab University College of Pharmacy, University of the Punjab, Lahore, Pakistan
[6]Department of Pharmacognosy, Faculty of Pharmacy, Federal Urdu University of Arts, Science and Technology, Karachi, Pakistan

Abstract: *Hertia intermedia* is a traditional medicinal plant of Balochistan, used for pain management and stomach problems. Current research work was intended to evaluate the anti-inflammatory and analgesic activities of crude ethanolic extract of *H. intermedia.* Anti-inflammatory activity was determined by the carrageenan-induced and histamine–induce Rat paw edema in rats, analgesic activity was determined by acetic acid-Induced writhing test, formalin-induced hind paw licking in mice and Tail immersion test. *H. intermedia* crude ethanolic extract showed significant ($p<0.05$) effect in both carrageenan and histamine-induced rat paw edema at both 250 and 500 mg/kg oral doses. There were significant analgesic activities in comparison with standard drug and control ($p<0.05$). It is concluded that *H. intermedia* crude ethanolic extract possesses significant anti-inflammatory and analgesic effects. However further studies may be carried out to isolate the phytochemicals responsible for anti-inflammatory and analgesic activities.

Keywords: Analgesic activity, anti-inflammatory, arthritis, *Hertia intermedia*.

INTRODUCTION

Among autoimmune disorders, rheumatoid arthritis is the commonest. In Arthritis, numerous inflammatory mediators are released, that are characterized by pain, inflammation of joint, loss of function, destruction of joint and if not treated may lead to permanent deformity (Srivastava *et al.,* 2012). Scientists are unable to find out the etiology of this disease (Mubashir *et al.,* 2014). The exact pathophysiology is still unknown, but the release of certain free radicals such as nitrous oxide and superoxide radicals are generated as by-products of cellular metabolism. The release of such free radicals can induce the production of interleukins (IL) and tumor necrosis factor (TNF-α) from T cells, which ultimately affect the production of cytokines, growth factors and adhesive molecules on immune cells, since such factors can cause tissue destruction and inflammation (Mukhopadhyay *et al.,* 2019, Choudhary *et al.,* 2015). Despite significant development in the management of arthritis by drugs and non-steroidal anti-inflammatory drugs (NSAIDs), exploration for other new drugs from natural sources has continued, because the synthetic drugs have numerous disadvantages and limitations (Baranwal *et al.,* 2012).

In ancient records, about five hundred (500) medicinal herbs have been reported for the treatment of arthritis, however scientific data is available on only few plants (Gautam *et al.,* 2020). There are some herbs that produce beneficial effects for chronic inflammatory conditions, such as osteo and rheumatoid arthritis. Secondary metabolites obtained from plants are important economical sources of new drugs (Mubashir *et al.,* 2014). Currently, many medicinal herbs have been reported with therapeutic properties for treatment of arthritis (Baranwal *et al.,* 2020). Hence, current study was carried out to evaluate the anti-inflammatory, anti-arthritic and analgesic activities of *Hertia intermedia* ethanolic extract.

Hertia intermedia belongs to Composite (also called Asteracea) family. In Balochistan, the plant is found in Quetta, Killa Abdullah, Chaman, Khanozai and Pishin (Anwar *et al.,* 2020). Locally this plant is called Manguli. It is used to relieve pain, stomach discomforts, headache and abdominal cramps during menstrual cycle (Tareen *et al,* 2010, Anwar *et al.,* 2020). Chemical constituents such as terpenoids, tannins, steroids, coumarins, flavonoids, and carbohydrates have been reported from the plant (Samiullah *et al.,* 2015).

MATERIALS AND METHODS

Collection of plant material

H. intermedia whole plant was collected from Hanna urak area of Quetta (longitude 67.02^0 and latitude 30.36^0). The plant was then authenticated by Botanist, Botany, Department, Balochistan University, Quetta, specimen

*Corresponding author: e-mail: pharmacognosist59@yahoo.com

with voucher number MY-235 was submitted for further reference.

Animals

Albino (Swiss) mice weighing about 25-30g and albino rats (Wistar) weighing about 150-180g were acquired from Dow university Karachi (Pakistan) for the investigational purpose. Plastic cages were used to keep the animals, standard conditions were maintained at standard temperature (20 to 23±1°C) and 12h light and 12h dark cycles. Standard (pellet) feed diet were administered. Animals have been allowed to acclimatize for seven (07) days at animal house with access to water and pellet diet.

Extraction of plant material

Plant material was dried, powdered and macerated with ethanol and solvent was vaporized using rotary evaporator at controlled pressure.

Preliminary phytochemical tests

Presence of different chemical compounds such as tannins, alkaloids, glycosides, steroids, saponins, terpenoids, coumarins, and falvonoids were determined by using standard tests and procedures (Jalalpure *et al.,* 2011).

Acute toxicity test

For this test, four groups of rats were used and each group consist of six (6) rats. Distilled water 5 ml/kg were administered to group I (Control), while the treatment groups received *H. intermedia* ethanolic extract at 175, 200, 500 mg/Kg body weight (PO). Animals received the extract daily for 14 days (Pariyani *et al.,* 2015).

Anti-inflammatory activities

Carrageenan-induced paw edema in rats

For this test, four (4) groups of rats were used and each group consist of 6 rats. 1% Carrageenan (0.2mL) w/v, was injected subcutaneously in right hind paw of the rats to induce edema. The paw diameter was measured by using a digital Vernier caliper before carrageenan injection and then diameter was measured hourly five (5) times, then after 24 and 48h. Normal saline (3mL/kg body weight) was administered to control group. Second and third groups were treated with 250 and 500mg/kg *H. intermedia* ethanol extract orally. Diclofenac sodium (Hisun Laborites) at the dose of 50mg/kg orally was given to standard drug treated group (Fourth group). Carrageenan was injected after one hour of saline, standard drug and 250 and 500mg/kg *H. intermedia* ethanol extract (Azza *et al.,* 2015).

Histamine -induced rat paw edema

To perform this test, four (4) groups of rats were used and each group was consist of six (6) rats. Distilled water 10 mL/kg was administered to control group orally and 250 and 500 mg/kg of *H. intermedia* ethanolic extract was administered to 2nd and 3rd group. Diclofenac (50mg/kg, p.o.) was administered to standard drug treated group, Edema of rat paw was measured after one hour of oral administration. Solution of histamine (0.001mg/mL) was injected in right hind paw of the rat to induce edema and edema was measured by using digital Vernier caliper. The paw thickness was measured at 1h intercession for 5h, after histamine injection (Kumar *et al.,* 204).

Analgesic activity

Acetic acid-induced writhing test

Analgesic activity was performed on mice by using acetic acid induced abdominal constriction test. Twenty (20) mice were grouped in four groups (5 mice in each group). First group was treated with distilled water and *H. intermedia* 250 and 500mg /kg was administered to 2nd and 3rd groups respectively and standard drug diclofenac sodium (50mg/kg b/w) was administered to 4th group. Acetic acid 1% (10mL/kg body weight) was injected intraperitoneally, after half an hour of drug and vehicle treatments, sum of writhes were noted for thirty minutes (Ilmi *et al.,* 2021).

Formalin-induced hind paw licking in mice

In this test, twenty (20) mice were used, mice were grouped in 4 groups (5 mice in each group). In this test 2.5% of formalin solution (20µL) subcutaneously injected underneath right hind paw surface. Licking of injected paw was considered as pain induction. Early nociceptive reaction usually occurs after five (05) min after formalin injection (first phase) and second phase starts after 15-30 minutes, 2nd phase represents the neurologic and inflammatory pain. Normal saline, 10mL/kg were administered to control group. Group 02 and 03 received *H. intermedia* 250 and 500mg/kg ethanol extract respectively. Standard drug treated group received 50mg/kg diclofenac sodium. The responses were recorded for first and second phases (Mondal *et al.,* 2021).

Tail immersion test

In this test, twenty (20) mice were used and divided into 4 groups (5 mice/group) i.e control group (vehicle treated), *H. intermedia* 250 and 500mg/kg (b/w) treated groups and standard drug treated group (diclofenac sodium 50mg/kg b/w), After each group treatment, tip of the tail of mice (last 1-2 cm) was immersed in the hot water (55±1) °C and time (seconds) were noted to withdraw the tip from hot water. The cut-off time was set at 15s to avoid injury. After 30 min of drug and extract administration flick response was determined (Aziz *et al.,* 2019).

Ethical approval

The ethical approval for animal use was granted by the Experimental Ethics Committee (No:FOPHS/AE/Pharm-08/2021, dated 07/09/2021) on Animal Use of the Faculty of Pharmacy and Health Sciences, University of Balochistan, Quetta

STATISTICAL ANALYSIS

Results were expressed as the mean ± SEM (standard error of mean). The one-way ANOVA test was followed by post-hoc Dunnett's test for multiple comparison by using SPSS software (Version 22), $p<0.05$ considered significant and $p<0.01$ considered highly significant (Ilmi *et al.,* 2021).

RESULTS

Phytochemical tests

In preliminary phytochemical tests, the presence of tannins, saponins, steroids, falvonoids, and triterpenoids were confirmed (table 1).

Acute toxicity test

In acute toxicity test, there was no mortality up to the dose of 5 g/kg (table 2).

Table 1: Phytochemical tests of *H. intermedia* crude ethanolic extract

S No.	Test	Result
1	Glycosides	Negative
2	Saponins	Positive
3	Tannins	Positive
4	Steroids	Positive
5	Flavonoids	Positive
5	Triterpenoids	Positive
6	Alkaloids	Negative

Table 2: Acute toxicity of *H . intermedia* crude ethanolic extract on rats

S No	Treatment	No of deaths
1	Control	Nil
2	*H. intermedia* 175mg/kg	Nil
3	*H. intermedia* 200mg/kg	Nil
4	*H. intermedia* 500mg/kg	Nil

N=06,

Table 3: Effect of *H. intermedia* crude ethanolic extract on body weight of rats

S No	Treatment	Body Weight (Mean±SEM)
1	Control	202.76±0.95
2	*H. intermedia* 175mg/kg	202.24±0.76
3	*H. intermedia* 200mg/kg	202.2±1.16
4	*H. intermedia* 500mg/kg	201.36±0.63

N=06, values are expressed as Mean ± SEM

Cage side observation

After administration of *H. intermedia* extracts, the treated groups of rats did not showed any abnormal behavioural, motor and neuronal dysfunctions and mortality The observations of eyes, fur, skin and interactive pattern like posture, gait, ANS and CNS activities of the treated rats were remained unaffected, in comparison with the control group. Results revealed that, oral LD50 of *H. intermedia* ethanolic extract was above than 500 mg/Kg body weight.

Body weight measurement

There was no significant variation in the body weight of the treated rats, in comparison with the control group (table 3). The Food water intake of the rats was consistent and regular during the treatment period (table 4 and 5).

Carrageenan-induced paw edema in rats

After administration of *H. intermedia* 250mg and 500mg oral dose, edema was significantly ($p<0.05$) decreased in comparison with diclofenac treated group. The intradermal administration of carrageenan on rat hind paw, produced topical and quick inflammation, determined paw volume was detected after 3h. The administration of *H. intermedia* 250mg, 500mg and diclofenac (50mg/kg) expressively repressed the carrageenan-induced inflammation. Significant ($P<0.05$) decrease in edema was observed after 60 min of plant extract administration. This dose also inhibited the second phase of inflammation with a maximum effect at the 5th hour (table 6).

Table 4: Effect of *H. intermedia* crude ethanolic extract on Food Intake of rats

S No	Treatments	g/rat/day (Mean ± SEM)
1	Control	172.02±0.69
2	*H. intermedia* 175mg/kg	171.64±1.79
3	*H. intermedia* 200mg/kg	170.84±1.71
4	*H. intermedia* 500mg/kg	168.65±1.05

N=06, values are expressed as Mean ±SEM

Table 5: Effect of *H. intermedia* crude ethanolic extract on Water Intake of rats

S. No	Treatments	ml/rat/day (Mean±SEM)
1	Control	73.48±0.71
2	*H. intermedia* 175mg/kg	75.7±0.73
3	*H. intermedia* 200mg/kg	78.46±0.85
4	*H. intermedia* 500mg/kg	80.04±0.33

N=06, values are expressed as Mean ±SEM

Histamine -induced rat paw edema

In this study *H. intermedia* 250 and 500 mg effectively suppressed the edema produced by histamine as compared with standard drug (table 7).

Analgesic activities

Writhing Test

In writhing test mean number of writhes for control group was 91.60±0.55, for *H. intermedia* 250mg/kg treated group 77.48±0.93, for 500mg/kg treated group it was 63.64±0.84 and for standard drug treated group it was 47.12±0.63. In this test plant produced significant effects ($p<0.05$), number of writhes were decreased at both doses

i.e. 250 and 500mg/kg oral doses in comparison with control and standard drug produced highly significant ($p<0.01$) results (table 8).

Formalin induced inflammatory pain

This method is most common method to determine the inflammatory pain. In this test, for control group, numbers of licking and biting in first phase were 72.19±0.57, in second phase it was 83.44±0.76, for *H. intermedia* ethanolic extract 250 and 500mg/kg treated groups numbers of licking and biting in first phase were 64.83±0.33 and 62.03±0.82 and in 2nd phase it was 75.31±0.34 and 71.36±0.36 respectively. For standard drug treated group numbers of licking and biting in first phase were 52.69±0.73 and in 2nd phase it was 51.06±0.62. *H. intermedia* ethanolic extract produced significant ($p<0.05$) decrease in numbers of licking and biting in first and 2nd phases as compared with control and standard drug produced highly significant ($p<0.01$) results (table 9).

Tail immersion test

In tail immersion test, time (seconds) taken by mice to withdraw the tail for control group was 5.42±0.07s, for *H.*

Table 6: Effect of *H. intermedia* crude ethanolic extract on Carrageenan-induced paw edema in rats

Treatment	0 hours	1 hour	2 hours	3 hours	4 hours	5 hours	24 hours	48 hours
Control	8.92± 0.17	12.87± 0.16	14.19± 0.07	14.31± 0.03	15.10± 0.18	15.65± 0.11	12.98± 0.08	12.17± 0.07
H. intermedia 250mg/kg	9.14± 0.01*	11.90± 0.18*	12.76± 0.11*	13.17± 0.08*	13.61± 0.09*	13.43± 0.21*	11.97± 0.06*	11.59± 0.17*
H. intermedia 500mg/kg	9.33± 0.05*	11.59± 0.11*	12.46± 0.08*	12.83± 0.18*	12.25± 0.14*	13.43± 0.18*	10.86± 0.34*	10.76± 0.36*
Standard Drug Diclofenac Sodium 50mg/kg	9.22± 0.03**	10.984± 0.36**	11.11± 0.04**	11.61± 0.10**	11.37± 0.15**	11.23± 0.07**	10.37± 0.16**	10.13± 0.04**

Table 7: Effect of *H. intermedia* crude ethanolic extract on Histamine -Induced Rat Paw edema

Treatment	1 hours	2 hour	3 hours	4 hours	5 hours
Control	3.31±0.15	3.56±0.12	3.91±0.03	4.16±0.04	5.04±0.05
H. intermedia 250mg/kg	3.14±0.09*	2.90±0.05*	2.61±0.13*	2.58±0.06*	2.43±0.02*
H. intermedia 500mg/kg	2.94±0.04*	2.84±0.04*	2.74±0.03*	2.50±0.04*	2.44±1.10*
Standard Drug Diclofenac Sodium 50mg/kg	2.88±0.05**	2.76±0.07**	2.52±0.07**	2.33±0.03**	2.38±0.13**

N=06, values are expressed as Mean ± SEM, *= $p<0.05$, **= $p<0.01$

Table 8: Effect of *H. intermedia* crude ethanolic extract on Acetic acid induced writhing test on mice

S. No	Treatment	Number of writhes
1	Control	91.60±0.55
2	*H. intermedia* 250mg/kg	77.48±0.93*
3	*H. intermedia* 500mg/kg	63.64±0.84*
4	Standard Drug Diclofenac Sodium 50mg/kg	47.12±0.63**

Table 9: Effect of *H. intermedia* crude ethanolic extract in Formalin-induced hind paw licking in mice

S. No	Treatments	Number of licking and biting 1st Phase	Number of licking and biting 2nd Phase
1	Control	72.19±0.57	83.44±0.76
2	*H. intermedia* 250mg/kg	64.83±0.33*	75.31±0.34*
3	*H. intermedia* 500mg/kg	62.03±0.82*	71.36±0.36*
4	Standard Drug Diclofenac Sodium 50mg/kg	52.69±0.73**	51.06±0.62**

Table 10: Effect of *H. intermedia* crude ethanolic extract on Tail immersion test of mice

S. No	Treatments	Time (seconds)
1	Control	5.42±0.07
2	*H. intermedia* 250mg/kg	7.376±0.09*
3	*H. intermedia* 500mg/kg	9.46±0.10*
4	Standard Drug Diclofenac Sodium 50mg/kg	11.62±0.14**

N=05, values are expressed as Mean ± SEM, *= $p<0.05$, **= $p<0.01$

intermedia ethanolic extract 250 and 500 mg/kg treated group it was 7.376±0.09 and 9.46±0.10 and for standard drug treated group it was 11.62+0.14s. Plant extract produced significant ($p<0.05$) results as compared with control and standard drug (table 10).

DISCUSSION

Arthritis affects both adults and aging persons equally. The occurrence rate ranges from 0.3 and 1%. Drugs obtained from natural sources have significant effect with less side effects. Natural drugs can be used in combination synthetic drugs for the treatment of arthritis (Foyet *et al.,* 2021).

In our study, results show that crude ethanolic extract of *H. intermedia* produced significant anti-inflammatory properties. To the best of our knowledge, this is the first report on anti-inflammatory activity of *H. intermedia* collected from Balochistan.

Carrageenan-induced rat paw edema method is most commonly used method for investigation of new anti-inflammatory drugs. After administration of carrageenan, an acute inflammatory response, involves 3 diverse phases, that result in release of various mediators. In 1st phase, (1.5h), histamine and serotonin are released, whereas in 2nd phase (1.5-2.5h), bradykinins are released. The 3rd phase occurs between 2.5 and 6 h, and facilitated by prostaglandins release. Last phase is the peak process in inflammatory response (Foyet *et al.,* 2015, Uttra *et al.,* 2017, Posatska *et al.,* 2019). Significant ($p<0.05$) anti-inflammatory effects were produced by *H. intermedia* crude extract, Studies show that inhibition of proinflammatory cells results in anti-inflammatory response (Mubashir *et al.,* 2014). However the exact mechanism of suppressing inflammation is not known. In phytochemical tests the *H. intermedia* crude extract showed positive results for presence of flavonoid, therefore it is hypothesized the presence of flavonoids are responsible for anti-inflammatory effect. Previous studies have showed that flavonoids have anti-inflammatory potential (Chinnasamy *et al.,* 2019).

H. intermedia crude ethanolic extract showed significant analgesic effect in writhing test, formalin test and tail flick test. After administration of acetic acid (IP), various inflammatory mediators like serotonin, histamine substance P, Prostaglandins, and bradykinin are released. These mediators produces chemical-induced visceral pain that leads to constriction of abdominal muscles, forelimbs expansion and elongation of the body. The PGE and PGF_2a levels also enhanced. Increased level of PG enhances inflammatory pain (Yimer *et al.,* 2020). Significant analgesic produced by *H. intermedia* crude ethanolic extract in writhing test may attributed to the presence of flavonoids. Previous reports show that flavonoids exert analgesic effect by increasing the serotonin (endogenous) concentration or interact with serotonin 2A receptor (5-HT2A and5-HT3) (Das *et al.,* 2014). Therefore it is suggested that the analgesic effect might be because of reticence of inflammatory mediators by the flavonoids present in the plant.

Formalin test is sensitive, reliable and valid method for several categories of analgesic drugs and have a number of advantages compared with other analgesic models, it shows no or little restraint, unconstrained reflection of whole array of behavioral reactions and better similarity to clinical pain (Demsie *et al.,* 2019)

Formalin-induced pain has 2 phases. The first phase is neurogenic phase (0-5 min after formalin administration) and commonly recognized to a straight effect of the stimulus on primary nociceptive neurons (Bukhari *et al.,* 2016) which depends on neurotransmitters such as serotonin, histamine and initiation of TRPA1 receptors articulated by neurons (Martinez *et al.,* 2016, Qnais *et al.,* 2017). *H. intermedia* crude ethanolic extract produced significant analgesic effects in both phase of formalin test as compared with control and standard drugs. It is suggested that the *H. intermedia* crude ethanolic extract has both peripheral and central anti-nociceptive effects.

In tail immersion test *H. intermedia* crude ethanolic extract showed analgesic effect by enhancing the time as compared to control group. Centrally acting analgesic agents raise the pain onset of the mice towards pressure and heat (Fan *et al.,* 2014). Consequently, it is suggested that plant extract may have centrally acting analgesic effect. Findings of the current study shows that *H. intermedia* crude ethanolic extract might be a good natural substitute for pain management.

CONCULSION

Form this study, it can be concluded that *H. intermedia,* plant extract has potential anti-inflammatory and analgesic effects, however, further studies are also required to isolate active constituents and determine the activity on large number of animals.

REFERENCES

Anwar M, Khan J, Manan A, Khan N and Hakeem A (2020). Anti-inflammatory activity of extracts and fractions of *Hertia intermedia*. *Al-Nahrain. J. Sci*., **23**(2): 80-83.

Aziz M, Mehedi M, Akter M, Sajon SR, Mazumder K and Rana M (2019). *In vivo* and *in silico* evaluation of analgesic activity of *Lippia alba*. *Clin. Phytoscience*, **5**(1): 1-9.

Azza Z and Oudghiri M (2015). *In vivo* anti-inflammatory and antiarthritic activities of aqueous extracts from *Thymelaea hirsuta*. *Pharmacog. Res.,* **7**(2): 213-216.

Baranwal VK, Irchhaiya R and Alok S (2012). Anti-arthritic activity of some indigenous plants: A review. *Int. J. Pharm. Sci. Res.,* **3**(4): 981-986.

Bukhari IA, Gilani AH, Meo SA and Saeed A (2016). Analgesic, anti-inflammatory and anti-platelet activities of *Buddleja crispa*. *BMC Complement. Altern. Med.,* **16**(1): 1-7.

Chinnasamy V, Subramaniyan V, Chandiran S, Kayarohanam S, Kanniyan DC, Velaga, VSSR and Muhammad S (2019). Antiarthritic Activity of *Achyranthes aspera* on formaldehyde-induced arthritis in rats. *Open Access Maced. J. Med. Sci.,* **7**(17): 2709-2714.

Choudhary M, Kumar V, Malhotra H and Singh S (2015). Medicinal plants with potential anti-arthritic activity. *J. Intercult. Ethnopharmacol.*, **4**(2): 147-179

Das BK, Al-Amin MM, Russel SM, Kabir S, Bhattacherjee R and Hannan JMA (2014). Phyto-chemical screening and evaluation of analgesic activity of *Oroxylum indicum*. *Indian. J. Pharm. Sci.,* **76**(6): 571-575.

Demsie DG, Yimer EM, Berhe AH, Altaye BM and Berhe DF (2019). Anti-nociceptive and anti-inflammatory activities of crude root extract and solvent fractions of *Cucumis ficifolius* in mice model. *J. Pain. Res*., **12**: 1399-1409.

Fan SH, Ali NA and Basri DF (2014). Evaluation of analgesic activity of the methanol extract from the galls of *Quercus infectoria* (Olivier) in rats. *Evid. Based. Complement. Alternat. Med.,* Article ID 976764.

Foyet HS, Tsala DE, Bodo JZE, Carine AN, Heroyne LT and Oben EK (2015). Anti-inflammatory and anti-arthritic activity of a methanol extract from *Vitellaria paradoxa* stem bark. *Pharmacognosy. Res.,* **7**(4): 367-377.

Gautam RK, Sharma S, Sharma K and Goyal S (2020). Evaluation of comparative anti-arthritic Activity of traditionally well documented medicinal plants in rats. *Indian J. Pharm. Sci.,* **82**(5): 791-796.

Ilmi H, Pamungkas IR, Tumewu L, Hafid AF and Widyawaruyanti A (2021). Analgesic and antipyretic activities of ethyl acetate fraction tablet of *Andrographis paniculata* in animal models. *Evid-Based. Complement. Altern. Med.,* Article ID 8848797.

Jalalpure SS, Mandavkar YD, Khalure PR, Shinde GS, Shela PA and Shah AS (2011). Antiarthritic activity of various extracts of *Mesua ferrea* Linn. seed. *J. Ethnopharmacol.,* **138**(3): 700-704.

Kumar T and Jain V (2014). Antinociceptive and anti-inflammatory activities of *Bridelia retusa* methanolic fruit extract in experimental animals. *Sci. World. J.*, Article ID 890151.

Martinez R M, Zarpelon AC, Domiciano TP, Georgetti SR, Baracat MM, Moreira IC, Andrei CC, Verri WA and Casagrande R (2016). Antinociceptive effect of *Tephrosia sinapou* extract in the acetic acid, phenyl-p-benzoquinone, formalin and complete Freund's adjuvant models of overt pain-like behavior in mice. *Scientifica.*, Article ID 8656397.

Mondal M, Quispe C, Sarkar C, Bepari TC, Alam MJ, Saha S, Ray P, Rahim MA, Islam MT, Setzer WN and Salehi B (2021). Analgesic and anti-inflammatory potential of essential oil of *Eucalyptus camaldulensis* leaf: *In vivo* and *in silico* studies. *Nat. Prod. Commun.,* **16**(4): 1-16.

Mubashir K, Ganai BA, Ghazanfar K and Akbar S (2014). Evaluation of antiarthritic potential of methanolic extract of *Gentiana kurroo* Royle. *Arthritis*, Article ID 810615.

MukhopadhyayN, Sampath V, Pai S, Babu UV and Lobo R (2019). Antiarthritic medicinal plants: A review. *Res. J. Pharm. Tech.,* **12**(1): 375-381.

Pariyani R, Safinar I I, Azam AA, Abas F, Shaari K and Sulaiman MR (2015). Phytochemical screening and acute oral toxicity study of Java tea leaf extracts. *Biomed. Res. Int.* Article ID 742420.

Posatska NM, Grytsyk AR, Struk OA and Klymenko AO (2019). Study of anti-inflammatory activity of *Verbena officinalis* L. extracts on the models of carrageenic and formalin edemas. *J. Pharm. Innov*., **8**(11): 141-144

Qnais E, Bseiso Y, Wedyan M and Alkhateeb H (2017). Evaluation of analgesic activity of the methanol extract from the leaves of *Arum palaestinum* in mice and rats. *Biomed. Pharmacol. J.*, **10**(3): 1159-1166.

Samiullah, Khalid S, Tareen RB, Khan N, Aslam S, Alam F and Suleman S (2015). Study of phytochemistry and antioxidant activity of *Hertia intermedia* (Boiss.) flowers of Balochistan. *Lasbela, U. J. Sci. Techl.,* 92-100.

Srivastava S, Singh P, Jha K K, Mishra G, Srivastava S and Khosa RL (2012). Evaluation of anti-arthritic potential of the methanolic extract of the aerial parts of *Costus speciosus*. *J. Ayurveda. Integr. Med.,* **3**(4): 204-208.

Tareen RB, Bibi T, Khan MA, Ahmad M and Zafar M (2010). Indigenous knowledge of folk medicine by the women of Kalat and Khuzdar regions of Balochistan, Pakistan. *Pak. J. Bot.,* **42**(3): 1465-1485.

Tiwari RK, Chanda S, Singh M and Agarwal S (2021). Anti-inflammatory and anti-arthritic potential of standardized extract of *Clerodendrum serratum* (L.) Moon. *Front. Pharmacol.,* pp. 1-11.

Uttra AM and Hasan UH (2017). Anti-arthritic activity of aqueous-methanolic extract and various fractions of *Berberis orthobotrys* Bien ex Aitch. *BMC Complement. Altern. Med*., **17**(1): 1-16.

Yimer T, Birru EM, Adugna M, Geta M and Emiru YK (2020). Evaluation of analgesic and anti-inflammatory activities of 80% methanol root extract of *Echinops kebericho* M. (Asteraceae). *J. Inflamm. Res.,* **13**: 647-658.