

ANTI-INFLAMMATORY AND ANTI-DIARRHEIC ACTIVITY
OF *Isocarpha cubana* BLAKE

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Summary

Introduction: The common chamomile (*Isocarpha Cubana Blake*) is a plant commonly used by Cuban people. It has the same properties scientifically tested on the sweet chamomile (*Matricaria Recutita, L.*). A few studies have been related to it and none has demonstrated that the plant has the properties attributed to it. A preliminary pharmacognostic study dealt favorably with the possible similarities of both plants. It was the basis that determined the study of its pre-clinical pharmacological effects. **Materials and methods:** The anti-inflammatory action was evaluated by means of the model of induced inflammation by carragenin which values the acute effect. It was completed with the two other models of inflammation: that of granuloma to evaluate the chronic effect of the extract and that of auricular edema to evaluate the topic anti-inflammatory action of the cream. Taking into account the wide use of this plant by our population as an anti-diarrheic and digestive medication, its effect on the intestinal duct was also studied. **Results:** The fluid extract diminished the percentage of acute inflammation produced by carragenin in three dose levels, being this dose effect dependent and similar to that of indomethacin. In the same way, it inhibited significantly the production of granulomatous tissue in the model of chronic inflammation. However, the chamomile cream at 5 % was not able to diminish, in a topic way, the inflammatory effect produced by the croton oil. While studying its activity on the intestinal tract the extract acted out as an anti-diarrheic medication, being able to decrease the laxative effect of glycerin. **Conclusions:** The fluid extract of the common chamomile demonstrated to have acute and chronic anti-inflammatory action as well as anti-diarrheic effect while the chamomile cream at 5 % did not have anti-inflammatory effect in a topic way.

Key words: anti-inflammatory, *Isocarpha*, preclinical, anti-diarrheic.

Introduction

The common chamomile (*Isocarpha cubana* Blake) is a wild plant that grows in our fields. Our people use it under the belief that it has the same scientifically proved properties as those of the sweet (or German) chamomile (*Matricaria recutita*, L). A lot of people think of them as the same plant even when the cultivation of the latter turns out very difficult in our climate conditions.

Few studies have been carried out that demonstrate the properties attributed to the plant. A previous pharmacological study had also shown that common chamomile is rich in triterpenes and steroids. They both are important substances in the anti-inflammatory activity⁽¹⁾. It also has a capillary image very similar to that of the sweet chamomile. This fact brings about the possibility of a similarity between their actions and it's the base that determines this study's objective of pre-clinically evaluating its anti-diarrheic and anti-inflammatory effects.

Materials and Methods

The pharmacological evaluation of the plant's anti-inflammatory action was made by means of pre-clinical tests based on internationally-established norms (OECD, ISO) using the chronic and acute inflammation models for the fluid extract of the plant and the topic inflammation model for the 5% cream made out the extract. In addition, it was evaluated the action of the extract on the intestinal transit. The fluid extracts were elaborated from the dried plant, properly identified and treated following the established norms for its production. All the studied products were quality-tested (chemically and micro-biologically). This guaranteed the necessary requirements for its evaluation.

Evaluation of the acute anti-inflammatory action

The test was made using the oral way and by the method of acute inflammation produced by Carragenin^(2,3). Male albino rats were used weighing between 200 and 250g. Five groups gathering five animals each were created: a positive control group (Indomethacine 7 mg/kg), a negative control group (without treatment), and three groups with three dosage levels (fluid extract 125, 200 and 325 mg/kg). The distribution of the animals within the groups was at random. The doses of active principle had a relation of geometric proportion of 1.6. The volume of both rear legs were measured with the aid of a pletismometer (UGO Basile) at intervals of time of 2, 4 and 6 hours after the Carragenin had been administered. The following formula was used to calculate the percentage of inflammation for each group:

$$\text{Inflammation} = \frac{V_p \text{ c/i} - V_p \text{ s/i}}{V_p \text{ s/i}} \cdot 100$$

Where: $V_p \text{ c/i}$ = inflamed leg's volume
 $V_p \text{ s/i}$ = not inflamed leg's volume

Finally, the statistic differences among the groups were evaluated using a one-way classification variance analysis and the Student Newman Keuls test with a $p < 0.05$.

Evaluation of the chronic anti-inflammatory action

The test was made using the granuloma method by means of cotton-pellet implants in rats ^(4,5). Fifteen male Wistar rats were used weighing between 120 and 250 g. The animals were distributed in three experimental groups (n=5): a group with common chamomile fluid extract, a group with Dexamethasone and a third group without treatment. The treatment was administered twice a day (morning and afternoon) for seven days using Dexamethasone doses of 15 mg/kg (1mL) and 370 mg/Kg (0.5 mL) of chamomile fluid extract.

The results were processed taking into account descriptive parameters and a simple or double ANOVA as well as multiple comparison tests (Newman-Keuls) or the Kruskal-Wallis test and Dunn's not-parametric multiple comparison test were used to compare the groups.

Evaluation of topic anti-inflammatory action

To evaluate this anti-inflammatory action a 5% cream was elaborated out of the fluid extract of the plant and using a hydrophilic base. The percentage was determined taking into account the anti-inflammatory results obtained with the extract.

The essay was carried out on rats of OF₁ line, weighing between 25 and 30g. It was employed the method of the acute inflammation in rat's ears produced by Croton oil ⁽⁶⁾. Three random groups of animals were created: a group without treatment, another with common chamomile cream (5 %) and a third group with Bencidamine cream. The following formulas were employed to determine the inflammation differences between the two ears and the percentage of inflammation's inhibition:

$$\text{Inflammation \%} = \frac{T \times 100}{ST} - 100$$

Where: T= ears weight percentage
 ST= not treated ears equivalent

$$\text{Inhibition \%} = \frac{C - T}{C} \times 100$$

Where: C= Edema extension of control group
T= Edema extension of treated group

Evaluation of the action on the intestinal transit

The study was carried out using the small intestine transit model ^(4,7). Fifteen Wistar male rats distributed in three experimental groups (n=5) were used: a group treated with common chamomile fluid extract, another group with this fluid extract and Glycerin and a third group treated with Glycerin. The results were statistically processed as same method of chronic anti-inflammatory test.

Results

Table # 1: Effect of common chamomile extract on carragenin induced-paw edema

Group	% of inflammation (X ± d.s.)		
	2h	4h	6h
Control	36.55±8.98 b*	43.42±9.38 d	29.27±6.16 f
Indomethacine 7 mg/kg	21.52±2.87 a	25.45±4.98 c	12.89±2.20 e
Extract 125 mg/Kg	26.06±3.99 a	32.09±4.44 c	17.33±6.43 e
Extract 200 mg/Kg	23.46±5.80 a	26.58±5.77 c	16.81±7.36 e
Extract 325 mg/Kg	22.89±5.93 a	24.92±10.97 c	11.81±2.24 e

* Different letters indicate significant differences (p<0.05)

Table # 2: Effect of common chamomile extract on granuloma formation induced by cotton-pellet implants

Treatment	Granuloma's weigh (M ± DS)(g)
Common chamomile fluid extract 370 mg/kg	0.090 ± 0.002 a
Dexamethasone 15 mg/kg	0.070 ± 0.002 b
No treatment	0.120 ± 0.001 c

Different letters indicate significant differences between groups (p<0.05)

Table # 3: Effect of common chamomile 5 % cream on inflammation in rat's ears produced by Croton oil

Group	Weigh differences among the ears (mg)		Inhibition %
	Left (control)	Right (treated)	
Croton oil	7.35 ± 0.74	11.21 ± 1.45a	-
Common chamomile cream 5%	6.39 ± 0.66	10.83 ± 1.08a	-
Bencidamine cream	6.80 ± 0.63	8.44 ± 1.14b	55.67

Different letters indicate significant differences between groups (p<0.05)

Table # 4: Effect of common chamomile extract on small intestine transit.

Treatment	Dose	Intestinal lengths (cm)		Intestinal Transit %
		Total (media ± ds)	Transited (media ± ds)	
Extract	1.0 mL	113.0 ± 7.2	57.4 ± 13.3	50.3 ± 10.6 a
Glycerin	1.0 mL	111.6 ± 8.8	88.8 ± 4.6	79.7 ± 3.9 b
Glycerin + Extract	1 capsule	117.2 ± 13.9	55.4 ± 4.7	41.94 ± 13.73 a

Different letters indicate significant differences (p<0.05)

Discussion

Evaluation of the acute anti-inflammatory action

Both indomethacine and the product object of study differ on the negative control at all times, being capable of reducing the inflammation on the rat's leg provoked by Carragenin with no remarkable differences them. The percentage of inflammation diminished proportionally to the increase of the product's dose, but it isn't statistically significant. The fluid extract of the common chamomile is an effective anti-inflammatory in the dose and model applied.

Evaluation of the chronic anti-inflammatory action

The daily administration of 370 mg/kg of the *Isocarpha cubana* Blake's fluid extract inhibited the formation of granulomatose tissue showing anti-inflammatory effect by reducing the weight of the granuloma. The analysis of results showed that exist statistically significant differences (p<0.05) between the treated groups. The results of acute and chronic anti-inflammatory tests are according to the presence of triterpenes and steroids in the plant showed in the previous studies ⁽¹⁾ and confirm the traditional use and the similarity of the capilar images of the common chamomile and the sweet chamomile.

Evaluation of the topic anti-inflammatory action

The best effects during this test were on the bencidamine-treated animals. The animals treated with common chamomile cream 5% had similar results to those not treated. That means the cream tested couldn't reduce the inflammatory effects on the rat's ear produced by Croton's oil.

Evaluation of the action upon the intestinal transit

The analysis of the results showed statistically remarkable differences (p<0.05) between the glycerin-treated groups and the fluid extract of the common chamomile. The product acts as an anti-diarrheic being even capable of counteracting the laxative effects of glycerin and confirming the traditional use of the plant.

Conclusions

The fluid extract of the common chamomile showed its acute/chronic anti-inflammatory action and anti-diarrheic effects on the pre-clinic models used. The common chamomile cream (5%) didn't show anti-inflammatory action in topic form.

Acknowledgments

To the bachelor Vladimir Molina, Maura Molina, Jillian Mora, Carlos Pineda and Rita Entralgo for their valuable and usefull help.

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