



UNIVERSIDAD DE GUAYAQUIL
DEPARTMENT OF CHEMICAL SCIENCES

Ciudadela Universitaria "Dr. Salvador Allende"
Telephone: 2293680, E-mail: fcquimic@ug.edu.ec
Guayaquil, Ecuador

FINAL REPORT

CODE: 38/05

TITLE:

Establishment of the potential anti-inflammatory effect of the product known as **Quina**, originating from NutraMedix Laboratories, LLC, Florida

OBJECTIVE:

To study the possible anti-inflammatory effect of QUINA, measured by edemas, induced by carragenin in the feet of laboratory mice.

BACKGROUND:

The method of inducing edemas by applying carragenin to the feet of mice is a classic model for the study of products with anti-inflammatory activity. The by-products of the metabolism of araquidonic acid via cecloxygenesis and the production of reactive species of oxygen are also involved. It is reported that there are four principle phases to this edema: an initial phase in which histamine and serotonin is released; a second phase measured by kinins; a third phase (about 5 hours) in which prostaglandins are liberated; and a fourth phase related to the local infiltration of neutrophils and their activation. This model has recently been recommended as very useful for the evaluation of anti-oxidant products with anti-inflammatory properties and free radicals of oxygen inhibitors.

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As discussed in numerous international works, the pharmacological study of the above-mentioned effect is indispensable, and guarantees (within the margin of error associated with the technique) that the potential for producing anti-inflammatory effects in humans will be learned.

The basis of this work is the pharmacological effect as an anti-inflammatory, as described in international literature (1, 2).

TECHNICAL, SCIENTIFIC AND SOCIOECONOMIC BENEFITS:

The demonstration of this product as an anti-inflammatory is important due to its potential as a new, plant-based medication, with its associated low toxicity. This was demonstrated by us in a previous work, allowing us to enter the product as a new medication in the appropriate Register.

VARIABLES TO MEASURE:

1. Weight of the feet
2. Percentage of Inflammation
3. Percentage of inhibition

PROCEDURES TO FOLLOW:

TEST MATERIALS:

Quina: The procedure followed was that described by CYTED (1996) and the Gerhard Voegel (1997).

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CHANGES IN THE CURRICULUM:

Changes did not take place in protocol proposed to the Unity of Quality Guarantee, whose number is referred to on page 1.

DATA FROM THE SAMPLE:

Organization soliciting services: NutraMedix Laboratories, LLC.

Person in charge of the Organization's application: Ing. Jose Icaza

Date of application: 7/15/05

Organization that carried out the work: University of Guayaquil, Department of Chemical Sciences.

Address: Ciudadela Universitaria "Dr. Salvador Allende"

Person in charge in the Executor Organization: Dr. Diadelis Ramirez Figueredo

Storage: The product was stored at room temperature with controlled access.

Form of presentation of the product: amber glass drop bottle containing 30 milliliters

Storage: The product was maintained at room temperature before and during the experiment, and as indicated was protected from light and kept in a locked cabinet.

INFORMATION WITH RESPECT TO THE HANDLING:

No special handling instructions were needed.

COMPOSITION OF THE PRODUCT:

Quina bark extract (Cinchona calisaya)

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EXPERIMENTAL PROCEDURE:

INTRODUCTION:

This experiment was carried out with the intention of determining the possible anti-inflammatory effect of QUINA, using oral application and employing the method of edema induced by carragenin as the inflammatory agent.

DOSAGE USED IN THE TEST:

In this study 1 mL of Quina per 25 grams of body weight was administered according to the dosage recommended by the manufacturer.

PRINCIPAL TEST:

METHODS AND TECHNIQUES:

Study Material: Quina

Animal Model: A single rodent species (mouse) was utilized, with a minimum of 5 animals of a single sex in each group. In this case, male mice with an average weight within $\pm 20\%$ (3), belonging to the Swiss line and coming from the Chemistry Department of the University of Guayaquil were appropriate and were utilized in the experiment.

The animals were maintained in quarantine conditions and were acclimated according to established procedures (4, 5), said period having a duration of five days minimum.

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Access to the water and the food was "ad libitum." (6, 7)

The animals were randomly distributed from within the different groups. (8)

Food was denied 4 hours before exposure to the test material.

The experiment lasted 6 days (5 of acclimation and 1 of test)

DEVELOPMENT OF THE METHOD:

The following four groups were constructed for the test:

TEST GROUPS	
1	Flebogenous agent Carragenin 1%
3	Carragin 1% + Quina 1 ml/25 grams

The mice were denied food for four hours then weighed, after which began the experiment.

The irritant solution of 1% carragenin was dissolved in physiological saline and 0.1 ml was administered on the right sole. The saline alone was administered to the other foot as a negative control.

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The composite solutions were administered orally one hour before the carragenin application. Five hours after the application of carragenin, the animals are euthanized in a saturated ether atmosphere, and their feet are cut at the knee and weighed.

RESULTS CALCULATIONS:

Outcomes are rated by calculating the weight of each mouse's feet, both the treated and untreated.

Percentage of inflammation:

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

Where T is the average of the weights of the treated feet (right) and ST is the average of the weights of the untreated feet.

Percentage of inhibition:

$$\% \text{ Inhibition of inflammation} = 100 - \left(\frac{\text{mean values of the treatment group}}{\text{mean values of the control group}} \right) \times 100$$

DESCRIPTION OF THE DOSAGE, METHOD OF ADMINISTRATION AND DURATION OF THE TEST:

The test was achieved by following the method established by CYTED and using the dosage indicated for each mouse.

The Quina was administered orally

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The Quina and the widely used pharmaceutical anti-inflammatory indomethycine were applied orally one hour before the application of the carragenin.

ANALITICAL RESULTS:

The results of the average value of the weights of the treated feet, and the standard deviations, are found in Table #1:

Group	Weigh of feet (g) (Mean \pm st. dev.)	% of inflammation	% of inhibition
Carragenina 1%	0.3 \pm 0.6a	96.1	---
Carragenina +Quina	0.18 \pm 1.0b	38%	40%
Carragenina + Indomethycine 10mg/kg	0.20 \pm 1.0c	53%	35%

Statistical significance between (a) with respect to (b) and (c) ($p < .05$)

As is demonstrated in Table # 1, Quina showed a statistically significant anti-inflammatory effect with respect to the control group with carragenin and superior to the referenced widely used non-steroidal pharmaceutical indomethycine. These results indicate the potential for this product as an anti-inflammatory.

This result marks guidelines for the investigation of the mechanism of action of this product as a potent anti-inflammatory for popular use.

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CONCLUSIONS:

Was demonstrated that QUINA has an anti-inflammatory effect.

GENERAL CONCLUSIONS:

Quina was demonstrated to have anti-inflammatory effect similar to indomethycine and can be used to address inflammation from inflammatory agents such as carragenine, as observed in animal testing and as appears in specialized literature.

PERSONNEL RESPONSIBLE FOR THE STUDY:

Responsible Professional:

Dr. Diadelis Ramirez Figueredo

Date: 07/26/05

Signature:

BIBLIOGRAPHY:

1. CYTED course for Researchers in the Discovery of new medicines, Lima November 1996, Edema Earpiece pp 83.
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8. Procedure. Euthanesia
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