# NEW ASPECTS IN QUALITY CONTROL OF "PALO AMARGO" (*Aeschrium crenata* Velloso- Simaroubaceae)

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Abstract

Different aspects on quality control of extracts from "palo amargo" (*Aeschrium crenata* Velloso -Simaroubaceae-) are being considered. The results suggest that preparation of solid pharmaceutical forms from hydroalcoholic extracts treated with lactose and povidone K30 is a simple procedure to stabilize them.

Studies on stability and quality control proposed allow the establishment of strategies in order to improve pharmaceutical preparations for pharmacological and biodisponibility analysis.

## 1. Introduction

Medicinal, flavour and aromatic plants are extensively used in the pharmaceutical, cosmetic and alimentary industries. One of the most traditional preparations for using them are based in the extraction with ethanol. However, these preparations show some problems when being used:

- Many extracts have lipofílic substances, which do not dissolve in water.
- Due to their complex composition, the stability of the extracts can be drastically damaged. Autooxidation of substances or hydrolysis are some of the involved mechanisms in the loss of pharmacological properties.

The correct determination of the doses.

In order to give a solution for these problems, hydroalcoholic extracts from *Aeschrion crenata* Velloso - Simaroubaceae - were prepared. This plant is known under the common name "palo amargo" in our country. The wood of this species is employed to obtain bitter principles (Vitagliano *et al.*, 1972 and Rosella *et al.*, 1991) and the tincture as pediculicide. Actually, its antiviral and insecticide properties are being tested.

The aims of this study are:

Transformation of liquid extracts from *Aeschrion crenata* Velloso -Simaroubaceae- in solid pharmaceutical preparations, more stable and easier to be administrated.

Establishment of methods to perform quality control of vegetal material and of the new preparations.

## 2. Materials and methods

### 2.1. Material

#### 2.1.1. Vegetal material

Commercial pieces of wood from "palo amargo" (*Aeschrion crenata* Velloso - Simaroubaceae-).

2.1.2. Adsorbents and/or complex

Povidone $K_{30}$.
Lactose.

2.2. Methods

2.2.1. Extract obtention

300 g of wood in pieces were macerated with 2000 ml alcohol 96° for a week, with periodical agitation.
The alcoholic solution was filtered throw paper Whatman N° 3.
The liquid was concentrated at 100°C in a bath. It was reduced until 98 ± 0.5% of the original volume.
The new extract was filtered using a nylon membrane of 0.45 μm.

2.2.2. Granulate obtention

In order to obtain the solid pharmaceutical preparation (granulate), two procedures were performed:

2.2.2.1. Aspersion of the concentrated extract on lactose

25 ml of the concentrated extract was added intermittently to 12 g de lactose, with agitation and drying with hot air after each application.

2.2.2.2. Granulation of the concentrated extract with povidone K 30 and lactose

30 ml of the concentrated extract was granulated with 12 g povidone K 30 and 66 g de lactose. The preparation was dried on a dish paced in an oven at 40°C during 24 hs, moving the granulate on the dish. To adjust the granules size a knife-mill was used.

2.2.3. Stability assays

2.2.3.1. Direct irradiation

Samples were placed without any protection at 15 cm under 254 nm UV- light during 24 hs.

2.2.3.2. Temperature action

Dried extracts were placed in Petri dishes without lid in ovens:
a) at 37°C ± 5°C, for 35 days and for three months,
b) at 60°C ± 5°C, for 72 hours and for 1 month.

2.2.3.3. Stability at room temperature

Dried extracts were placed on Petri dishes without lid, and light-protected at room temperature for 12 months.

2.2.4. Pharmacobotanic quality control of vegetal material

2.2.4.1. Macroscopic study

2.2.4.2. Boodle's disgrigation (10% KOH at 100°C for 10 min. and 25% chromic acid at room temperature for 30 min.).

2.2.4.3. Obtention of slices

2.2.4.4. Staining procedure: with 2% floroglucine alcohol 96° and exposition to fumes of concentrated HCl.

2.2.5. Granulate quality control

2.2.5.1. Planar liquid chromatography (Wagner *et al.*, 1996)

Thin layer chromatography (TLC) and high resolution thin layer chromatography (HPTLC) were performed on silica gel 60 F254 plates.
Solvent: chloroform-methanol (95:5).
Placed quantity: 3µl of a 60% methanolic solution of vegetal extract.

Detection:
a). Observation under UV-light at 254 nm and 365 nm.
b). Reaction with vainilline/ sulfuric acid, examination at daylight.
Quassine (Rf: 0.70), paraine (Rf: 0.30) and isoparaine (Rf: 0.40).

2.2.5.2. High resolution liquid chromatography (HPLC) (Nestler *et al.* 1980, Robins *et al.* 1984)

Solvent: Water: methanol: acetonitrile: tetrahydrofurane 95:10:10:7.
Flux: 1,0 ml/min.
Detection: 257 nm.
Column: RP-8 Licrospher 250-4 Endcapped.
Sample: 20 µl.
Quassine (RT: 18.72 min), paraine (RT: 27.67 min) and isoparaine (RT: 32.09 min).

## 3. Results

3.1. Pharmacobotanic quality control

3.1.1. Macroscopic study

"Palo amargo" wood appears as pale yellow chips of variable size. Fragments of black-brownish bark are found too.

3.1.2. Disgrigation

Vessels: short and thick, with vestuled pits.
Fibers: main fiber-tracheids 250-350 µm long, with vestuled pits.

3.1.3. Slices

Slices agree with those from Literature (O'Donnell, 1937).

3.1.4. Granulate obtention

By both methods solid water-soluble preparations from the alcoholic extract of *A. crenata* were obtained.

3.1.5. Studies on stability of *A. crenata* granulates

The relative concentration of each quassinosid from granulates corresponding to the different treatments were compared with those present in a fresh hyroalcoholic extract.

## 4. Discussion

Both methods are based on improving solubility by means of two mechanisms:
* increasing of contact surface and interchange between solid and solvent,
* obtention of soluble complexes.

The procedure described as "aspersion" is the example for the first of these mechanisms, when covering excipient particles (lactose) with the extract.

Granulation has two advantages: the capacity of povidones to form soluble complexes with organic compounds and to stabilize them.

Lactose cannot avoid oxidative processes.
Thus, preparation of solid pharmaceutical preparations from alcoholic liposoluble extracts (either by complex formation or by adsorption) provides a simple and practical way to solubilizate extracts and obtain an easy controllable pharmaceutical preparation.

Studies on stability and quality control proposed allow the establishment of strategies in order to improve pharmaceutical preparations for pharmacological and biodisponibility analysis.

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Table 1 - Studies on stability of *A. crenata* granulates

| **Treatment** | **Complex** | ***Quasine*** | ***Paraine*** | ***Isoparaine*** |
|---|---|---|---|---|
| **Direct Irradiation** | Lactose | +/- | +/- | +/- |
| | Lactosa/povidoneK30 | + | + | + |
| **Temperature: 37°C** 35 days | Lactose | + | + | + |
| | Lactose/povidone K30 | + | + | + |
| 3 months | Lactose | - | - | - |
| | Lactosa/povidone K30 | +/- | +/- | +/- |
| **Temperature: 60°C** 72 hours | Lactose | - | - | - |
| | Lactose/povidone K30 | + | +/- | +/- |
| 1 month | Lactosa | - | - | - |
| | Lactosa/povidone K30 | ? | - | - |
| **Stability at room temperature** 1 year | Lactose | - | - | - |
| | Lactose/povidone K30 | ? | - | - |

+: detected in same concentration +/-: detected in less concentration
?: detected only in traces -: not detected
Hydroalcoholic extracts begin to exhibit damage 15 days after elaboration.