

## QUALITATIVE ASSESSMENT OF CHEMICAL CONSTITUENTS OF *PICRIS BABYLONICA*

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### ABSTRACT

The objective of the present study was to assess the phytochemical constituents like; tannins, saponins, flavonoids, steroids, alkaloids, anthraquinones, and cardiac glycosides. The qualitative analyses were performed by the well-known protocols of both chemical and physical tests (including Infrared (IR) spectroscopy) available in the literature. The current study revealed that *Picris babylonica* is characterized chemically by the presence of different secondary metabolites such as flavonoids, saponins, triterpenoids/steroids, tannins and carbohydrates at different levels in different extracts of different plant organs and the absence of anthraquinones, cardiac glycosides and alkaloids in all parts of plant under investigation.

**KEYWORDS:** *Picris babylonica*, *Asteraceae*, phytochemical and IR.

### 1. INTRODUCTION

Family *Asteraceae* is one of the largest families of flowering plants and contains around 22,000 species, distributed into 1620 genera of different plant types.<sup>[1]</sup> The genus *Picris* comprises around 40 species which are perennial plants that grow throughout the world.<sup>[2-3]</sup> Many species of *picris* investigated previously and revealed that they contain many and various types of constituents like sesquiterpenes, phenolic compounds, flavonoids and terpenoids.<sup>[2-8]</sup> *Picris babylonica* is distributed in many parts of Saudi Arabia. According the recent literature review using most relevant resources, *P. babylonica* was investigated for its

volatile constituents only.<sup>[9]</sup> These findings provoked us to carry out this study to stand on the phytochemical contents in various plant parts.

## 2. MATERIALS AND METHODS

### 2.1. Plant Material

*P. babylonica* was collected from Riyadh region, Saudi Arabia. Different parts (leaves, stems, flowers and roots) of plant were separated and subjected to air-drying according to universal standard herbarium procedures. A voucher sample was kept in Department of Pharmaceutical Sciences, College of Clinical Pharmacy, King Faisal University.

### 2.2. Extraction and fractionation of different plant organs extracts

The powdered air dried plant material [leaves, stems, flowers and roots of *P. babylonica*, 600, 300, 150 and 150 g respectively] were exhaustively extracted twice at room temperature (each for 3 days) using 3 L 70% methanol in water through cold maceration method at room temperature to avoid degradation of possible active constituents. The solvent was removed via distillation under reduced pressure using rotary evaporator and resultant residues were directly freeze-dried to yield the total extracts of leaves, stems, flowers and roots weighting 36, 16, 5.5 and 9 g respectively, which were stored in freezer for the next steps.

Exactly 36, 16, 5.5 and 9 g of leaves, stems, flowers and roots total extracts respectively were suspended in distilled deionized water (200 ml) using a separating funnel and partitioned with n-hexane (6×500 ml). The resulting n-hexane phases were combined to be concentrated to the least amount using rotary evaporator and then dried to give 17, 6, 1.5 and 0.5 g respectively then were stored in a deep freezer in well-closed container. The remaining aqueous part was subjected to partition with chloroform (4×500 ml). The obtained chloroform fractions were also combined and its amount was reduced to the minimal amount through using rotary evaporator and then freeze dried to give 5, 2, 0.5 and 2 g, respectively, and then all were put in fridge in a strong-tight container for later use.

Similarly, the ethyl acetate and n-butanol extracts were also developed using the same above-mentioned procedure to give 7, 2.5, 1 and 2.5 g respectively for ethyl acetate fraction and to give 2, 1.5, 0.5 and 1 g respectively for n-butanol fraction. The remaining aqueous fraction was also freeze-dried to powder to give 4, 3, 1.5 and 2 g respectively and kept for further use in the freezer in an air-tight container.<sup>[10]</sup>

### 2.3. Phytochemical Screening of different plant organs extracts

Chemical testes were carried out on the n-hexane, chloroform, ethyl acetate, butanol and aqueous extracts of different organs using standard procedures to identify the nature of its constituents.<sup>[11-13]</sup>

#### 2.3.1. Test for flavonoids

Part of dried extracts of each organ was boiled with 10 ml of distilled water for 5 min and filtered while hot. Few drops of 20% sodium hydroxide solution were added to 1 ml of the cooled filtrate. A change to yellow color, which on addition of acid changed to colorless solution, depicts the presence of flavonoids.

#### 2.3.2. Test for saponins

Part of dried extracts of each organ was separately boiled with 10 ml of distilled water in a bottle bath for 10 min. The mixture was filtered while hot and allowed to cool. Demonstration of frothing: 2.5 ml of filtrate was diluted to 10 ml with distilled water and shaken vigorously to form a stable persistent froth.

#### 2.3.3. Test for steroids and/or triterpenoids

Part of dried extracts of each organ was separately boiled with 10 ml of distilled water in a bottle bath for 10 min. The mixture was filtered while hot and allowed to cool. Five milliliters of each extract was mixed in 2 ml of chloroform. Three milliliters of concentrated H<sub>2</sub>SO<sub>4</sub> was then added to form a layer. A reddish brown precipitate coloration at the interface formed indicated the presence of steroids and/or triterpenoids.

#### 2.3.4. Test for alkaloids

Part of dried extracts of each specimen was separately boiled with water and 10 ml hydrochloric acid on a water bath and filtered. The pH of the filtrate was adjusted with ammonia to about 6-7. A very small quantity of Dragendorff's reagent (potassium iodide 0.11 M, bismuth nitrate 0.6 M in acetic acid 3.5 M), the test tubes were observed for orange to brown turbidity.

#### 2.3.5. Test for anthraquinones

Part of dried extracts of each organ was boiled with 2 ml of 10% hydrochloric acid for 5 min. The mixture was filtered while hot and filtrate was allowed to cool. The cooled filtrate was partitioned against equal volume of chloroform and the chloroform layer was transferred into

a clean dry test tube using a clean pipette. Equal volume of 10% ammonia solution was added into the chloroform layer, shaken and allowed to separate. The separated aqueous layer was observed for any color change; delicate rose pink color showed the presence of an anthraquinone.

#### **2.3.6. Test for tannins**

Part of each dried extracts of each organ was separately boiled with 20 ml distilled water for 5 min in a water bath and was filtered while hot. One milliliter of cool filtrate was distilled to 5 ml with distilled water and a few drops (2-3) of 10% ferric chloride were observed for any formation of precipitates and any color change. A bluish-black or brownish-green precipitate indicated the presence of tannins.

#### **2.3.7. Test for cardiac glycosides**

Part of dried extracts of each organ was separately boiled with 10 ml of distilled water in a bottle bath for 10 min. The mixture was filtered while hot and allowed to cool. Five milliliters of each extract was treated with 2 ml of glacial acetic acid containing one drop of 10% ferric chloride solution. This was underplayed with 1 ml of concentrated  $H_2SO_4$ . A brown ring at the interface indicated the deoxy-sugar characteristics of cardenolides. A violet ring may appear below the ring while in the acetic acid layer, a greenish ring may be formed.

#### **2.3.8. Test for carbohydrates**

Part of various extracts were dissolved separately in 4 ml of distilled water and filtered. The filtrate was treated with 2-3 drops of alcoholic alpha-naphthol and 2 ml of concentrated  $H_2SO_4$  was added along the sides of the test tube. Appearance of brownish violet ring at the junction of the two liquids indicates the presence of carbohydrates.

### **2.4. Infra-red spectroscopy screening of different plant organs extracts**

Infra-red spectra of the different plant organs extracts were recorded to detect the presence of various functional groups of different constituents as a confirmatory tool for their occurrence. One  $\mu$ l of different plant organs extracts was loaded on KBr disc and placed in the sample chamber of FT-IR spectrophotometer and the spectra were recorded in the range of 4000–500  $cm^{-1}$  on Shimadzu 330 FTIR spectrometer. The most important absorption frequencies appeared in functional group region as well as fingerprint region of the spectra.<sup>[14]</sup>

### 3. RESULTS AND DISCUSSION

#### 3.1. Phytochemical screening of different plant organs extracts

The preliminary phytochemical screening of extracts of different plant organs of *P. babylonica*, showed the presence of various secondary metabolites such as flavonoids, saponins, triterpenoids/steroids, , tannins and carbohydrates at different levels in different fractions of plant organs and the absence of anthraquinones, cardiac glycosides and alkaloids as shown in Table 1.

**Table1: Preliminary phytochemical screening of different extracts from different plant organs.**

| Plant organ |    | Flavono<br>ids | Saponi<br>ns | Triterpenoid<br>s/steroids | Alkal<br>oids | Anthraq<br>uinones. | Tanni<br>ns | Cardiac<br>glycosides | Carbohyd<br>rates |
|-------------|----|----------------|--------------|----------------------------|---------------|---------------------|-------------|-----------------------|-------------------|
| Leaves      | nH | -              | +            | +                          | -             | -                   | -           | -                     | -                 |
|             | CH | +              | +            | -                          | -             | -                   | -           | -                     | -                 |
|             | EA | ++             | +            | +                          | -             | -                   | +           | -                     | +                 |
|             | BT | +              | +            | +                          | -             | -                   | +           | -                     | ++                |
|             | AQ | +              | +            | -                          | -             | -                   | -           | -                     | ++                |
| Stems       | nH | -              | +            | +                          | -             | -                   | -           | -                     | -                 |
|             | CH | +              | +            | -                          | -             | -                   | -           | -                     | +                 |
|             | EA | +              | +            | +                          | -             | -                   | +           | -                     | +                 |
|             | BT | +              | +            | -                          | -             | -                   | -           | -                     | +                 |
|             | AQ | -              | +            | -                          | -             | -                   | -           | -                     | +                 |
| Flowers     | nH | -              | +            | +                          | -             | -                   | -           | -                     | -                 |
|             | CH | +              | +            | -                          | -             | -                   | -           | -                     | +                 |
|             | EA | ++             | +            | +                          | -             | -                   | +           | -                     | +                 |
|             | BT | ++             | +            | +                          | -             | -                   | +           | -                     | +                 |
|             | AQ | +              | +            | -                          | -             | -                   | -           | -                     | +                 |
| Roots       | nH | -              | +            | +                          | -             | -                   | -           | -                     | -                 |
|             | CH | +              | +            | -                          | -             | -                   | -           | -                     | +                 |
|             | EA | +              | +            | -                          | -             | -                   | +           | -                     | +                 |
|             | BT | -              | +            | -                          | -             | -                   | -           | -                     | +                 |
|             | AQ | -              | +            | -                          | -             | -                   | -           | -                     | +                 |

nH, n-Hexane; CH, chloroform ; EA, Ethyl acetate; BT, Butanol; AQ, Aqueous. + (present); - (absent); ++ (present more concentration).

#### 3.2- Infra-red spectroscopy screening of different plant organs extracts

FT-IR spectral analyses data of different plant organs extracts revealed the presence of multiple functional groups. Spectral data of most of the extracts confirmed the presence of functional groups such as alcoholic, aldehydic, acidic and aromatic groups at different characteristic frequencies of IR radiation. These results also helped to confirm the observations from preliminary phytochemical screening.

#### 4. CONCLUSION

In the light of the results of this study it is very clear that different organs of *P. babylonica* contain variety of different constituents that have not been discovered yet. Hence, further research towards the isolation and identification of various constituents will be carried out in future.

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