

ORIGINAL ARTICLE

Phytochemical study of two medicinal plants (*Rosmarinus officinalis* and *Anthémis nobil*) from the Haddada region (El Tarf-Algeria)

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Medicinal plants are still the weak source of active principles known by their therapeutic properties, which leads us to the conservation of the biodiversity of local plant species. To this end we have carried out a phytochemical screening of two medicinal plants most used in the region of El Tarf. The present work was carried out in the laboratory of chemistry at the University of El Tarf in order to realize phytochemical analyses of some active principles of two medicinal plants, the Rosemary family of Lamiaceae (*Rosmarinus officinalis*) and the Chamomile family of Asteraceae (*Anthémis nobil*) are aromatic plants still used in the traditional medicine of many countries as antiseptic, antiviral, antispasmodic, stomatal, expectorant, in aromatic or in infusion. The results obtained show that the phytochemical screening revealed the presence of alkaloids, flavonoids, saponins, volatile oils, tannins and anthocyanins.

Keywords: *Rosmarinus officinalis*, *Anthemis nobili*, Medicinal plant, Active principles, Phytochemical screening.

Introduction

The history of aromatic and medicinal plants "A.M.P." is associated with the evolution of civilizations. In all regions of the world, the history of peoples shows that these plants have always occupied an important place in medicine, in the composition of perfumes and in culinary preparations (Belkacem, S, 2009). Herbal medicine is one of the oldest medicines in the world. It represents an interesting alternative to treat and cure without creating new diseases. Despite the phenomenal development of the pharmaceutical and chemical industry, popular interest in herbal medicine has never stopped evolving (Benguerba, A, 2008). Nowadays these two types of medication are found intimately linked since the molecular model of most of the drugs put on the market, have their origin in the plant (Ladu, G., Cubaiu, L., d'Hallewin, G., Pintore, G., Petretto, G.L., Venditti, T, 2015). In developing countries, between 70 and 95% of the population uses medicinal plants for primary care due to lack of access to prescribed drugs but also because plants have been shown to be really effective. It is estimated that at least 25% of all modern medicines are derived directly or indirectly from plants, through the application of modern technologies to traditional knowledge. (Sahi, L, 2016). In addition, the side effects induced by the drugs worry the users. They are turning to less aggressive treatments. It is estimated that 10 to 20% of hospitalizations are due to side effects of chemical drugs. (Benhouhou, S, 2015). Medicinal plants contain a large number of active

molecules of multiple interests put to good use in industry, food, cosmetology and in dermatology. (Menaceur, F, 2012). Among these molecules are coumarins, alkaloids, phenolic acids, tannins, lignans, terpenes and flavonoids (Benkiki, N, 2006). Flavonoids potentially possess biological, anti-inflammatory, anti-carcinogenic, antimicrobial and antioxidant activities. (Santos, R.D., Shetty, K., DaSilva Miglioranza, L.H, 2014). In Algeria the use of medicinal plants occupies an important place in the life of the population given the floristic wealth of our country. (Zakariène, G., Rokaitytė, A., Ramonaitė S, 2015). Given this richness of the plant world, our work is interested in medicinal plants most used in the region of el tarf, including chamomile (*Anthémis Nobili*) and rosemary (*Rosmarinus Officinalis* L).to develop an experimental method in the laboratory allowing a phytochemical study of some active ingredients of our two plants. The choice of two of these plants is based on the one hand on the importance of its family "Lamiaceae" and on the other hand on its known and frequent traditional use among the populations studied.

Materials and Methods

Plant material

The studied species collected on a determined area in the same regions (Haddada) the forest of Haddada constitutes the prolongation of the wooded littoral zone of the extreme North-East of Algeria. It is limited to the North by Segleb, to the East by the Tunisian borders, to the West by the commune of Souarekh and to the South by the commune of El Aioun.

The following Table 1 shows the dates and locations of harvest.

Table 1. Sampling characteristics.

Species	Date of harvest	Location of harvest	Part harvested
Rosemary	17/04/2021	Haddada	Leaf, stem
Chamomile	17/04/2021	Haddada	Flower
Rosemary	25/04/2021	Haddada	Leaf, stem
Chamomile	25/04/2021	Haddada	Flower

Drying

The harvested plants must be dried in the shade, in well-ventilated places, protected from humidity and water, the drying control being done daily. The maximum temperature allowed for the drying of plants is 30 (Boumaza, D, 2011) and (Bruneton, J, 2009).

The conservation

After drying, the best way to preserve the dried plants is to place them in a thick original bag and store them away from light and humidity. In these conditions of conservation, the plants can keep all their properties until their analysis in the laboratory. (Mansour, S, 2015) and (Nogaret, A.S, 2003) and (. Solinas, V., Deiana, S., Gessa, C., Bazzoni, A., Loddo, D, 1996).

Methodologies

The plant materials that will allow us a "phytochemical screening" test are coarsely ground in an electric mill. The powder of this material is submitted to a set of phytochemical tests: alkaloid test, flavonoid test, tannins, steroids....etc.

Phytochemical screening

Preparation of the infusion

2 grams of the dry matter are covered with 20ml of very hot or boiling water, after 3 to 6 minutes, or stir slightly and filter (Fig. 1).



Fig. 1. Preparation of the infusion.

Volatile oils

Macerate 10 g of the leaf powder in 40ml of distilled water with constant stirring 30 ml extract is filtered. 2 ml of filtering medium is shaken with 0.1 ml of diluted NaOH and a small amount of diluted HCL. A white precipitate is formed with the volatile oils (Fig. 2).

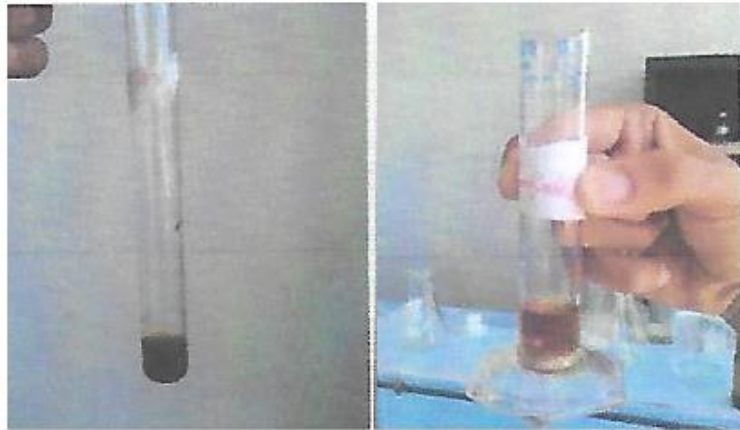


Fig. 2. Reveal volatile oils.

Tannins

A portion of the 10% infusion is diluted with distilled water in a ratio of (1/4) and 3 drops of ferric chloride $FeCl_3$ of 10% are added (Fig. 3).



Fig. 3. Reveal tannins.

Flavonoids

Macerate 10 g of the powder in 150 ml of HCL diluted at 1% for 24 hours, filter and proceed to the following test:

Take 10 ml of the filtrate, make it basic by adding NH_4OH ; the appearance of a light yellow color in the upper part of the test tube, indicates the presence of flavonoids (Fig. 4).

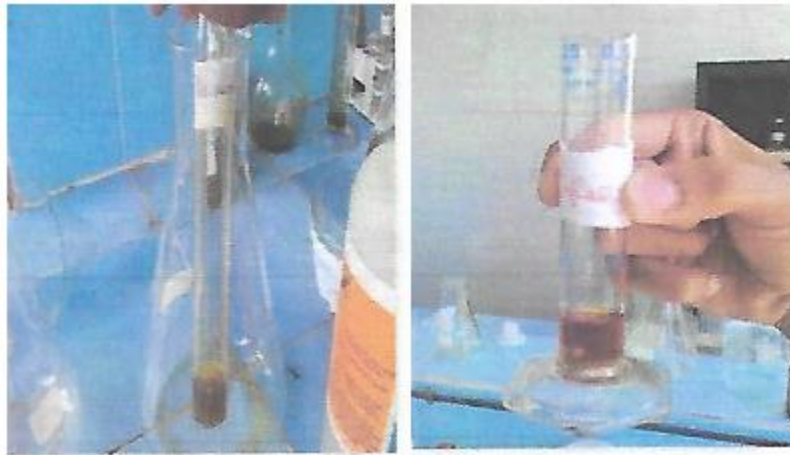


Fig. 4. Revealing flavonoids.

Alkaloids

Macerate 5 g of the powder in 50 ml of 10% HCL, filter then add to the filter a few drops of Mayer's reagent. A white precipitate indicates the presence of alkaloids (Fig. 5).



Fig. 5. Revealing alkaloids.

Anthocyanins

Rest on the change of color of the infusion at 10% with change of PH. We add to the infusion some drop of pure HCL, we have a change of color, and then we add some drop of NH_4OH . We have another change of color; this indicates the presence of anthocyanins (Fig. 6).



Fig. 6. Revealing Anthocyanins.

Leuco-anthocyanins

We heat 5 ml of the 10% infusion with 4 ml (ethanol/pure HCl 3/1) in a 50°C water bath for a few minutes. A cherry red color indicates the presence of leuco-anthocyanins.

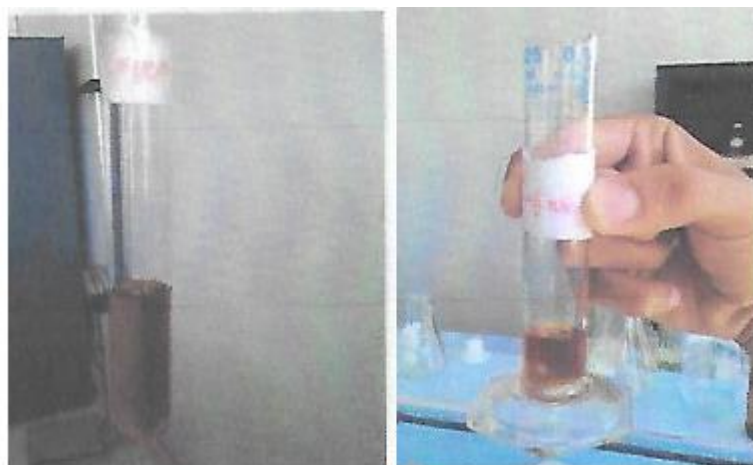


Fig. 7. Revealing leuco-anthocyanin.

Saponins

One gram of the dry powder is weighed into a flask into which 10 ml of distilled water is added and boiled for 5 minutes. The mixture is filtered; 2.5 ml of the filtrate is added to 10 ml of distilled water in a test tube. The test tube is shaken vigorously for about 30 seconds and then left to stand for half an hour. Foaming foam reveals the presence of saponins.

Coumarins

One gram of the material is placed in a tube, in the presence of a few drops of water, the tube is covered with paper soaked in diluted NaOH and brought to boil. Afterwards a drop of water is taken and put on TLC plate. Any yellow fluorescence shows the presence of coumarins after examination under UV2.

The cardinolides

Macerate 1 g of the powder in a 20 ml of distilled water and filter, take 10 ml of the filtering, extract it with a mixture of chloroform CHCl_3 and ethanol $\text{C}_2\text{H}_5\text{OH}$. Evaporate the organic phase and dissolve the precipitate in 3 ml of acetic acid CH_3COOH , add a few drops of FeCl_3 followed by 1 ml of concentrated H_2SO_4 on the wall of the test tube, the appearance of a green-blue color in the acid phase indicates the presence of cardinolides.

Terpenes and sterols

Macerate 1g of the powder in 20ml of ether for 24 hours, filtered then evaporated, the residue obtained is dissolved in anhydrous. The addition of pure sulfuric acid develops, in the presence of sterol or terpene products, a mauve coloration turning to green.

Results

The results obtained are grouped in the following Table 2 and 3:

Table 2. Results obtained from "phytochemical screening" tests of the rosemary plant.

Active ingredients	Specific reagents	Results
Saponins	NaOH	Positive
Volatile oils	NaOH+HCL	Positive
Tannins	FeCl_3	Positive
Coumarins	NaOH	Positive
Flavonoids	$\text{HCL}+\text{NH}_4\text{OH}$	Positive

Alkaloids	Mayer's reagents	Positive
Anthocyanins	HCL+NH ₄ OH	Positive
Leuco-anthocyanins	Ethanol	Positive
Terpenes and sterols	Ether	Positive
Free quinones	Petrol ether	Positive
Cardinolds	CHCl ₃ +H ₂ H ₅ OH+CH ₃ COOH+FeCl ₃ +H ₂ SO ₄	Positive

These analyses confirm the presence of active ingredients in the leaves of the rosemary plant.

Table 3. Results obtained from phytochemical screening tests of the chamomile plant.

Active ingredients	Specific reagents	Results
Saponins	NaOH	Negative
Volatile oils	NaOH+HCL	Positive
Tannins	FeCl ₃	Positive
Coumarins	NaOH	Positive
Flavonoids	HCL+NH ₄ OH	Positive
Alkaloids	Mayer's reagents	Positive
Anthocyanins	HCL+NH ₄ OH	Positive
Leuco-anthocyanins	Ethanol	Positive
Terpenes and sterols	Ether	Positive
Free quinones	Petrol ether	Positive
Cardinolds	CHCl ₃ +H ₂ H ₅ OH+CH ₃ COOH+FeCl ₃ +H ₂ SO ₄	Positive

These analyses confirm the presence of the active ingredients in the flowers of the Chamomile.

The positive test confirms the presence of active ingredients for both plants. Concerning the Rosemary the active ingredients are located in the leaves while the Chamomile contains several active ingredients in the flowers.

Discussion

Through the ethnobotanical study carried out among the population of the study area, it appears that there is a diversity of practices, as for the species, treated symptoms, parts used, doses of preparation and mode of use. There is also a diversity of information concerning the people surveyed; age class, profession, sex, family situation and level of education. (Rosua, A, 1987) and (O.M.S, 2000) and (Ozenda, P, 2004).

During our study in the laboratory we found that the active principles are present in both plants used, Rosemary is more used because of its availability throughout the year and the diversity of its therapeutic properties. (Ozenda, P, 2004) and (Satyal,A.A.P., Jones,T.H., Lopez,E.M., Mcfeeters,R.L., Awadh Ali, N.A., Mansi,I., Al-kaf,A.G., Setzer,W.N,2017).

It is noted that the chemical composition of medicinal plants is under the effect of several factors influenced on the absence, appearance and even distribution of active principles, such as water, soil and climatic conditions of the study area.

In spite of the development of the industry of the drugs of chemical origin, the traditional phytotherapy constitutes currently a source of remedy par excellence (PARK, H.J. and CHA H.C, 2003) and (Akrouit, A., Hajlaoui, H., Mighri, H., Najjaa, H., El Jani, H., Zaidi, S., Neffati, M., Essent J, 2010).

Natural products were and still are an inexhaustible source of complex and diverse structures given the role that some pure compounds can play in a lot of applications, namely pharmaceutical industry, food industry, cosmetic industry, perfumery, etc. (Penchev, P.I, 2010) and (Rasooli, I., Fakoor, M.H., Yadegarinia, D., Gachkar, L., Allameh, A. and Rezaei, M.B, 2008) and (Ndjouondo, G.P., Ngene, J.P., Ngoule, C.C., Kidik pouka, M.K., Ndjib, R.C., Dibong, S.D., and Mpondo, E, 2015). In order to enhance the value of medicinal plants used in traditional medicine, it is necessary to know them first, to study them to confirm their therapeutic effects.

The most used parts of these plants are leaves, stems and flowers.

Conclusion

The phytochemical study of our plants: Rosemary of the Lamiaceae family, and Chamomile of the Compositae family, which are the subject of our work, chosen on the basis of its wide use in traditional medicine in our region, allowed us to establish a phytochemical screening, which consists in detecting the active principles existing in the studied part of the plant by means of coloring and precipitation reactions using specific reagents. The chemical study of our plants was not limited to what has been presented in this work, because the complete study is very vast, and what has been studied is only a part of this study.

The medicinal plants are still the weak source of active principles known by their therapeutic properties, which leads us to the conservation of the biodiversity of local plant species.

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