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Medicinal plants in floristic regions of El Harrouch and Azzaba (Skikda-Algeria): Production and therapeutic effects

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ABSTRACT

Our study on the production and the therapeutic effects of medicinal plants used by the local population of the two study areas of El Harrouch and Azzaba in the region of Skikda (Algeria), shows that the species are divided into 22 species and 14 families. The most used is the Lamiaceae family with the species *Th. vulgaris* with hight consumption. The study of botanical characters of International Union for the Protection of New Varieties of Plants (UPOV), agronomic (installation condition of culture) and biochemical (essential oil, total phenols and flavonoids) of this plant demonstrates their economic and therapeutic interest. According to the results, it was found that the installation of thyme culture is very important, and knowledge of installation conditions and botanical characteristics (as UPOV recommendations), are very important for better adapt this beneficial culture through their therapeutic and commercial value at the Skikda region.

KEYWORDS: Medicinal plants, production, therapeutic, El Harrouch, Azzaba, Th. vulgaris

INTRODUCTION

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The medicinal plants and remedies are important treatment strategies since time immemorial, which led to maintain a living therapeutic tradition known since our ancestors [1]. Algeria for its floristic richness included more than 600 species of medicinal plants. According to several researches, the use of traditional medicine in our country has since seen a great consumption compared to other past years. While the cultivation of medicinal plants has always remained limited by some areas and some plants only. On the other hand, most medicinal plants are harvested in the mountains or in areas where they grow spontaneously without human intervention [2]. One of the singularities of plants is to form many compounds which do not enter into the general metabolism but which originates from its products; these are the secondary metabolites, are the main tools of the coevolution plantliving being and environment[3]. They are found in the organs of the plant (root, stem, leaf, flower, fruit and seed); and they are generally localized to cellular tissue levels: vacuoles (nicotine, glycosides, sapnosides, tannins, anthocyanins), constituents of the walls (lignin) and isolated secretory hairs, secretory sacs, and secretory channels (essential oils) [4]. Tubocuraraine; the most powerful muscle relaxant is derived from curare (Chondrodendron tomentosum) and cocaine used as anesthetics; is taken from coca (Erythroxylum coca) [5]. Today, plant-based treatments are coming back to the fore as the effectiveness of drugs such as antibiotics (considered as the near-universal solution to serious infections) is decreasing, bacteria and viruses have gradually adapted to drugs and resist them more and more. Herbal medicine based on natural remedies is well accepted by the body, and often associated with conventional treatments. It is now experiencing an exceptional renewal in the West, especially in the treatment of chronic diseases such as asthma or arthritis [6]. A medicinal plant only becomes useful after having undergone a number of transformations that seek to release its active ingredients and make them available to the body. Because of this, to remove these active ingredients from a herbal drug. Most often, liquid is used to dissolve them [7]. Among the most available preparations, we find: The simple solution, maceration, infusion, digestion (or infusion in the water bath), decoction and leaching (or percolation). The objective of our work is to carry out ethnobotanical survey with herbalists in two regions of Skikda, Azzaba and EL-Harrouch to evaluate the importance of the medicinal plants used by the local population.

MATERIALS AND METHODS

Study Area

The Skikda Province covers an area of $4,137.68 \text{ km}^2$ with 130 km of coastline. Our study is carried out on two Daïras of the

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Province of Skikda, the Daïra of El Harrouch and the Daïra of Azzaba. This choice is based on the knowledge of the two regions and the permanent use of the local population of medicinal plants sold at the level of herbalists. The Daïra d'Azzaba: it covers an area of 35.5 km², La Daïra El Harrouch: is located in the south of Skikda province. Its global surface area is 101.80 km².

The climate of the Skikda region is of Mediterranean type, characterized by two seasons: a mild and rainy winter, and a hot and dry summer. The recorded annual precipitation varies between 600 and 800 mm/year, the months of July and August are the driest months [8]. The temperatures recorded are of the order of 9 °C in winter and 27 °C in summer, which gives a cold winter and a hot summer, characteristics of the Mediterranean climate. The winds are generally north-east and south-west. Humidity generally during the day is around 70%, except for the month of January when it slightly exceeds this threshold. The soil of the Skikda region is loamy-sandy, silty-clayey and sandy-clay.

Study Methods

Ethnobotanical survey was carried out in order to highlight the importance and the therapeutic use of the ten most used medicinal plants by the local population, to classify them in order of use, in order to know the plant most used in the two regions of study (Azzaba and El Harrouch), to realize the installation of the culture. The ranking of medicinal plants according to their degree of consumption, ie the most sold at the level of herbalists.

Progress of the Survey

Data on the use of medicinal plants were collected from 13 herbalists practicing at El Harrouch (08 herbalists) and Azzaba (05 herbalists), with the completion of 10 questionnaires/ herbalist, a total of 130 questionnaires.

Model of the ethnobotanical survey card

Study of the Most used Medicinal Plant

For the leaves twenty plants with 3 repetitions were selected, making a total of 60 leaves. Regarding the flowers five (05) plants were selected with 4 repetitions, for a total of 20 flowers. Observations on the leaf should be made on fully developed leaves in the middle third of the plant. Observations on the flower must be made at the time of the full flowering of the variety. Observations on open flowers should be made on the first day of opening.

Biochemical Study of the Plant

The plant material used in this study is *Th. vulgaris*, which grows in the region of Azzaba (Commune of Djendel) in the Skikda province. The harvest is carried out in a reference site far from any disturbance and impact of pollution likely to disturb the chemical composition of the plant. The part used is the aerial part, and the sample is dried in the shade far from any source of artificial heat.

Extraction of Essential Oils

Extraction of the essential oils of *Th. vulgaris* was accomplished using a Clevenger type aparatus. 50 g of the dry plant cut into small pieces with 1000 ml of tap water are added to the flask for 3 h, the oil is separated from the hydrolate by decantation with using the magnesium sulphate (MgSO 4) in order to eliminate traces of water.

Extraction of phenols and flavonoids

5 g of the dry plant (leaves and stems) were macerated during 24h in 100 ml of 80% hydroalcoholic solvent which is heated to boiling, all thereafter is filtered on Whatman paper. The maceration is repeated three times with renewal of the solvent. The solvent is removed from the filtrate by rotavapor.

Ethnobotanical fact sheet					
Profile of the herbalist:					
N °: Date: Municipality: Age: Sex: Level of study: Origin:					
Ranking of the best-selling plants Medicinal plants used N ° 1					
Scientific name of the plant and the family:					
Vernacular name of the plant:					
Arabic name of the plant:					
Amazigh name of the plant:					
Origin of the plant: 🗆 Spontaneous 🗆 Cultivated 🗆 Imported					
Treated disease: 🗆 Breathing apparatus 🗆 Digestive system 🗆 Circulatory system 🗆 Genitalia 🗆 Dermal 🗆 Analgesic					
Preparation: 🗆 Infusion 🗆 Decoction 🗆 Powder 🗆 Fumigation 🗆 Maceration 🗆 Bkhour (essence) 🗆 Syrup 🗆 ointment 🗆 inhalation					
\Box Lotion \Box Suppository \Box Spray					
Part of the plant used: 🗆 root 🗆 stem 🗆 leaf 🗆 flower 🗆 inflorescence 🗆 fruit 🗆 whole plant					
How to use the plant: \Box dry in the shade \Box cool \Box dry in the light					
Dose of the plant: $\Box < 50g \Box 50g \Box 100g \Box 100g$					
Processing time: □ 1 day □ [2d-5d] □ [6d-10d] □ [11d-15d] □ 15d					
Use of the plant alone or associate: 🗆 Only 🗆 Associate with what					
Origin of the information: 🗆 Reading 🗆 Achabe 🗆 Pharmacist 🗖 Experience of others					
Price (DA / Kg) : \Box 100 \Box [100-500] \Box [500-10,000] \Box [10,000-50,000] \Box 50,000					

Table 1: Creation of a description of *Th. vulgaris* according to the recommendations of UPOV. [9]

N	Caractère	Expression	Echelle
01(*)	Plant: port	Dressed port /Half erect/Horizontal	1/3/5
02(*)	Plant: height (cm)	Very low /Low/Average/high/Very high	1/3/5/7/9
03(*)	Plant: diameter (cm)	Small/Average/great	3/5/7
04(*)	Foliage: density	Small/Average/Strong	3/5/7
05(*)	Stem: length (cm)	Short/Average/Longue	3/5/7
06	Stem: thickness (cm)	Thin/Average/thick	3/5/7
07(*)	Stem: distribution of leaves	Only at the base/Only in the middle/	1/2/
		Only at the top/All along the stem	3/4
08(*)	Stem: position of inflorescences	At the top/Along the upper quarter/	1/2/
		Along the top half/Along the top 2/3/	3/4/
		All along the stem	5
09	Stem: flower density	Low/Average/Strong	3/5/7
10(*)	Leaf: form	Elliptical/Oval/Rhomboid	1/2/3
11(*)	Leaf: length (cm)	Small/Average/great	3/5/7
12(*)	Leaf: width at the basal part (cm)	Small/Average/great	3/5/7
13	Leaf: length / width ratio (cm)	Small/Average/great	3/5/7
14(*)	Leaf: variegation	Absence/Present	1/9
15(*)	Leaf: main color	Green yellow/green/Green-blue/Gray green	1/2/3/4
16(*)	Leaf: intensity of the main color	Clear/Average/dark	3/5/7
17(*)	Flower: size (cm)	Small/Average/great	3/5/7
18(*)	Flower: petal color	White or slightly pink/Pink/Mauve/Purple	1/2/3/4
19(*)	Time of beginning of flowering (date and duration)	Very early/precocious/Average/Late/Very late	1/3/5/7/9

(*): Mandatory characters for all varieties

Determination of Total Phenols Content (Tpc)

The content of phenolic compounds is evaluated according to the method described by Touati, [10]. The TPC of the extracts are determined with reference to a standard curve made with gallic acid (100 to 1,000 μ l/mL) and the results are expressed in mg of gallic acid per mg of extract (mg EAG/g) and in g EAG per kg of dry matter (g EAG/kg).

Determination of Flavonoids Content

The content of flavonoids is determined according to the method described by Djeridane et al. [11]. The flavonoid concentrations of the extracts are determined with reference to a standard curve made with quercetin (100 to 1,000 μ l/ml) and the results are expressed in mg equivalent quercetin per g of extract (mg EQ/g) with reference to a calibration curve.

RESULTS AND DISCUSSIONS

Interpreting Questionnaires from Ethnobotanical Survey Cards

According to the ethnobotany survey, 27 plants were found used by the local population of the two study areas (Table 2). According to the order of use, we note that the most used plant was Thyme with 10% and the least used plants are, Germander, Sagebrush, Ginkgo, Licorice, Henna, Rue, Linden, Nettle and Sorrel with only 0.76%. The ranking of the 27 plants used is influenced by the most common diseases and their therapeutic treatments in both study areas. For example the thyme is ranked first for its therapeutic treatment against diseases of the digestive and respiratory system, and cutaneous, oral and dental diseases. Then for the Mint the same thing, we find that the local population uses this plant with a percentage of 9.23% for the preparation of herbal teas against abdominal pain and respiratory problems. With regard to Fenugreek, the seeds of this plant are used as a fortifier, against anemia, stress and digestive problems, and help the production of milk maternal. The Anise is used with a percentage of 6.92 against digestive problems. While Chamomile plants, Fennel and Senna are used by 6.15% against inflammations of the digestive tract and as a sedative against stress. Clove, Verbena And cultivated Nigella are used with a percentage of 5.38%, to treat the problems of the digestive system, cough, and as an anti-inflammatory.

The plants used are divided into thirteen families. The most used family is Lamiaceae with a rate of 30% followed by Apiaceae with a rate of 16.92%. While the least used are Ginkgoaceae, Lythraceae, Malvaceae, Rutaceae and Tiliaceae with a rate of 0.77% (Figure 4). According to the Figure 3, the majority of medicinal plants are of cultivated and imported origin. On the other hand, Thyme, Mint, Chamomile and Anise are spontaneous plants with a frequency of 12, 4, 1 and 8 respectively.

According to the Figure 5, we notice that leaves and fruits are the most used organs in therapeutics. While in Thyme, Mint and Verbena, the leaves and stems together are the most used. From the Figure 6, we note that the most common and most used mode of use is the drying in the shade. While the consumption of the fresh plant is used slightly in certain species such as Thyme, Mint, Fenugreek, Senna, Verbena and Nigella cultivated. The price of the plants varies between 1,000 and 5,000 DA. Most of the plants are priced at 5,000 DA (Thyme, Mint, Senna, Clove, cultivated Nigella and Verbena). While Anise, Chamomile and Fennel their price is 1,000 DA.

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Table 2: The most used plants in both study areas

Ν°	Common name of the plant	Frequencies
1.	Thyme	13
2.	Mint	12
3.	Fenugreek	9
4.	Anise	9
5.	Roman Chamomile	8
6.	Fennel	8
7.	Sene	8
8.	Clove	7
9.	Verbena	7
10.	grow Nigelle	7
11.	Cress	6
12.	Ginger	6
13.	Cumin	5
14.	Sage	5
15.	Rosemary	4
16.	Lavender	3
17.	Turmeric	2
18.	Marjoram	2
19.	Germander	1
20.	Wormwood	1
21.	Ginkgo	1
22.	Licorice	1
23.	Henna	1
24.	Street	1
25.	Lime	1
26.	Nettle	1
27.	sorrel	1



Figure 1: Administrative map of Algeria and political map of Skikda province. The regions where this study was conducted have been highlighted in dark color

On the other hand Fenugreek represents a price of 500 DA (Figure 7).

Plant Cultivation

Needs of the plant

Soil: Limestone, sandy, stony soil, drained and dry soil; P^H of soil is alkaline, neutral minimum 6.5; Climate Adaptation: Mediterranean;Water requirement: once the crop is planted, it does not require water. However, at planting or in the following;



Figure 2: Percentage of the main plants used in the two study areas (El Harrouch and Azzaba)



Figure 3: Distribution of the 27 plants used in the two study areas by family



Figure 4: Distribution of plants according to their origin

weeks, climatic conditions may require one or two waterings to facilitate plant growth [12].

Multiplication Method

Sowing: Sow in March indoors or in May outdoors in nursery, without covering seeds that need light to germinate; Place the seeds on the surface and barely cover;Keep the soil slightly damp at first with an indoor sprayer;The seed takes between 15 and 21 days to rise;Repeat two months later, when the plants are developed;Dig a small hole and gently lay the plan;Tamp the soil and water slightly; Space plans 20 to 30 cm [12];In organic farming, simple rank is preferred for weeding issues;



Figure 5: Distribution of plants according to the parts of the plant used



Figure 6: Distribution of plants according to their mode of use

mechanical: single rows spaced 1.60 m apart; or 18,000 to 20,000 plants/ha;The spacing between rows may vary depending on the available equipment (tractors;griffins.). On the row, the distance between plants is about 25 cm [13].

Cutting

Practice cuttings in July-August. The rooting will be done after two months, you can set up your plants the following spring. The cut of thyme can be done in two ways:

In a box or pot

- Choose a beautiful and vigorous stem and proceed in the morning, when the plant is still wet at night.
- Cut frankly a stem of 10 to 15 cm, using a sharp pruner.
- Delete the flowers if there are leaves at the base of the stem leaving only the top leaves.
- Prepare a box or pot with a very draining mix (50% limestone garden soil, and 50% river sand or perlite).
- Make a hole with a pencil and insert the cuttings.
- Tighten the soil slightly around the stem and sprinkle gently.
- Place in the shade and sheltered from the wind [12].

In the water:

- For selection and cutting of the stem, proceed as for potato cuttings.
- Install the cutting in a glass of water. Place the glass in a bright place but not in direct sunlight.
- Roots will appear in a few days. Then plant in individual pots.



Figure 7: Plant distribution by price (in DA)



Figure 8: Herbarium of Th. vulgaris

- After watering the cuttings to ensure a good recovery. Until the appearance of signs of restarting the vegetation, the soil should not dry between two waterings.
- After the recovery, install in the ground, water then, when the plant is well established, cultivate as an adult subject [12].

Crop requirements of the plant

Work of the soil

Fall plowing and/or subsoiling to obtain a deep soil, favoring a good development of the root system in depth. The plowing is resumed very early in the winter with a harrow or a cultivator to allow a plantation in the best conditions (smooth ground, without clods).

If possible, at least two false seeding should be done to reduce the stock of bad soil seeds. Destruction of raised weeds will be mechanical [12].

Cultural rotation

It is necessary to respect a rotation of at least two years without thyme or other species of perennial Lamiaceae between two

Table 3: Creation of plant factsheets according to UPOV. [9] recommendations

N°	Character	Expression	Scale
01(*)	Plant: port	Half erect	3
02(*)	Plant: height (cm) tab 5	20.51cm	/
03(*)	Plant: diameter (cm) tab 5	0.19cm	/
04(*)	Foliage: density fig 26	Average	5
05(*)	Stem: length (cm) tab 5	18.57cm	/
06(*)	Stem: distribution of leaves fig 27	All along the stem	4
07(*)	Stem: position of inflorescences	Along the top 2/3	4
08(*)	Stem: flower density	Average	5
09	Leaf: form fig 27	Elliptical	1
10(*)	Leaf: length (cm) tab 5	2.67cm	/
11(*)	Leaf: width at the basal part (cm) tab 5	1.28cm	/
12	Leaf: length / width ratio (cm) tab 5	2.08cm	/
13(*)	Leaf: variegation fig 27	Absence	1
14(*)	Leaf: main color fig 26	green	2
15(*)	Leaf: intensity of the main color fig 27	dark	7
16(*)	Flower: size (cm) tab 5	0.53cm	/
17(*)	Flower: petal color	White or slightly pink	1
18(*)	Time of beginning of flowering (date and duration)	Precocious (date: mid-month of May, duration one week)	3

plantations. This precaution makes it possible to respect the natural fertility of the soil but also to clean the soil.

Favorable precedents are cereals and legumes, ideally two years of legumes, followed by a cereal to benefit from the nitrogen effect and clean the soil [12].

Fertilization

Contributions in the year of planting: According to the previous crop and as a rule, the crop does not fertilize the year of planting.

Contributions in production: through the use of organic fertilization and the amendment.

Organic

- a. Organic fertilization: about 40 units of N.P.K as organic fertilizer type 9-12-0, or 7-8-12 or 6-6-6.
- b. Organic amendment: 10 to 15 t/ha of lavender or lavender straw compost can be brought in the winter before planting. It is also possible to make an annual contribution of 5 t/ha of compost on installed culture, but this requires adapted equipment.

Since thyme is sensitive to chlorosis (yellowing symptoms generalized on the plot), it will sometimes be necessary to bring iron to the culture, in the form of iron chelate [12].

Irrigation

Once the culture is established, it does not require irrigation. However, at planting or in the following weeks, climatic conditions may require one or two waterings to encourage plant growth, especially in the case of herbaceous plants.

Thyme values water well

Plots for irrigation can be cut twice a year, but their life is reduced (4 years instead of 8 years) [12].

Harvesting Condition and Conservation

The harvest: no harvest in the first year. Harvesting takes place in early May, at the full bloom stage until the end of flowering. It is mechanized with two types of harvesters used: the selfloading mower and the lavender cutter.

Storage: Once the thyme is collected, it is dried immediately and kept in well-ventilated and shaded places away from sunlight. The preservation procedure is as follows:

- a. Wash the stems with water to remove the dust, without letting them soak.
- b. Drain them well.
- c. Bring the stems together in bunches.
- d. Hang them in dry and well-ventilated places, head down.
- e. When the set is dry enough, you can beat it to loosen the leaves.
- f. Place the leaves in airtight boxes [12].

Diseases and Pests

The most common pests are: defoliating caterpillars, noctuids, and the beetle (*Arima marginata*), which can cause damage as soon as vegetation starts in April, May, on young shoots. There may also be flea beetle attacks. In the presence of these pests (3 to 4 larvae per plant), the treatment must be carried out with an approved insecticide. An insecticide is registered and effective in organic farming. Sectorial diebacks are often observed on certain varieties: they correspond to attacks of soil fungi such as Pythium and Fusarium. In addition to varietal tolerance, it is therefore advisable to pay the greatest attention to crop rotation and proper functioning soil (compost intake) [12].

Biochemical Contents

The extraction of 50 g of leaves and stems of *Th. vulgaris* is given 01 ml of essential oil, a yield of 2%. This yield is very important compared to other species, which generally give a yield between 1.5 and 2%. This difference is marked by the fluctuations of the climatic, edaphic and anatomical factors of the glands of the essential oils (size and number). The results obtained confirm those obtained by AFNOR on the varieties of Lemon Tree, and also show the richness of this species in essential oil.

- Aspect: Moving Liquid
- Color: Pale yellow
- Odeur: Nice characteristic, kinetic, spicy.

The content of the phenolic compounds was 26.87 mg EAC/g dry extract of the plant (Table). These results show a very high *TPC* compared to the work of Zeghad. [14] which recorded a rate of 9.07 mg EAG/g. The quantity of phenolic compounds in the methanolic extracts of the plants studied depends on

essentially: from their origin [15], the variety, the growing season, the harvest season, the climatic and environmental conditions, the geographical location, the various diseases that can affect the plant, the maturity of the plant and shelf life [16].

The flavonoid content was 27.5 mg/g EQ for the dry extract of thyme (Table). These results show a rise in flavonoid levels compared to the results of Zeghad. [14] with 8.56 mg/g EQ. These differences in results are explained by fluctuations in the environment and heredity.

CONCLUSION

On the basis of 130 questionnaires carried out in the two study areas of El Harrouch and Azzaba, the floristic analysis of the listed species shows that the species used, are divided into 27 species and 14 botanical families, of which only one species belongs to the junction of Gymnosperms (Ginkgo biloba). The branch of Angiosperms is represented by 25 species that are distributed among 13 families. Of the 14 families encountered, only three families clearly dominate the use of medicinal plants by the local population with: 30% of Lamiaceae, 16.92% of Apiaceae and 13.85% of Fabaceae. This dominance is influenced by the wealth of families in chemical substances with therapeutic effect. The classification of plants according to their use shows that thyme is the most used plant with a rate of 10% followed by mint with 9.23%. This ranking is due to the main diseases that exist in the two study areas which shows that the local population suffers in the first place from digestive disorders. The analysis of the prices of the medicinal plants at the level of the herbalists show that the majority of the plants the price of a kilogram it exceeds 1,000 to 5,000 DA. From the results obtained, it was found that the installation of the thyme culture is very important in the two study areas. Therefore, the knowledge of the installation conditions and the botanical characteristics (according to the UPOV. [9] recommendations), are very important to better adapt this beneficial culture by their therapeutic and commercial interest in the region of Skikda. The biochemical analysis of thyme, concerning the essential oils, the total phenols and the flavonoids shows a richness and a remarkable diversity which confirms the therapeutic effect of this plant. Finally, this study shows the excessive use of the population by medicinal plants and opens the way to other agronomic, ethnobotanical, phytochemical and ethnopharmacological studies likely to lead to a scientific evaluation and validation leading, possibly to the elaboration new drug principles.

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