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EcoAgriTourism, in the light of its multidisciplinary character, is a wide-open journal which brings together the opinions of specialists from both academic and economic environment, fostering fruitful collaborations.

The journal's structure covers all aspects of the fields approached, the focus being on original and current researches with applications in agriculture, food industry and rural tourism.

Collaborators may feel free to undertake biological and technical aspects as well as aspects with social, cultural and environmental impact. Information of general interest is also welcome for the agriecology-food-tourism axis

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Brussels 2022 - RURAL PACT AND MOUNTAIN AREAS

The Rural Action Plan, which was adopted with the Long-term Vision for the EU's rural areas, represented the European Commission's commitment to acting for rural areas. The Rural Pact, which was part of the **Rural Action Plan**, aims to strengthen multi-level governance for the EU's rural areas. This means that it will engage the European, national, regional and local level, all of which have a key role to play in the future of rural areas, *to work together*.

Of course, a direction of great attractiveness for the rural and agro-forestry area in general is Responsible Tourism with its distinct activities of *Ecotourism*, *Rural Tourism* and especially *Agritourism*.

What is noteworthy is that the EU's upland regions also feature prominently in cohesion policy in the context of recovery and resilience. Without going into detail, in order to have a general idea of cohesion in the EU's mountain regions, we can mention the following **lines of action**, which can also be found in the field of Responsible Tourism:

- Jobs and economic growth in mountain regions;
- The socio-economic dimension of mountain regions;
- Environmental protection and combating climate change in mountain regions;
- Accessibility and connectivity in mountain regions.

The current and acute problems of mountain areas mean that only the transposition and customisation of policies at local level, with the development of *integrated development strategies*, can best respond to the specific needs of the mountains and local specificities.

Director of the publication,

Prof. Romulus GRUIA, PhD, PhD supervisor, Scientific Leader

SUMMARY

Edito	rial: Brussels 2022 - RURAL PACT AND MOUNTAIN AREAS - GRUIA Romulus
5	PROPEERTIES OF WOOD BIOMASS FROM COMPOSITE MATERIALS GH. C. SPIRCHEZ, A. LUNGULEASA, L. GACEU
9	PLANT COVER ANALYSIS AND DIACHRONIC EVOLUTION USING GIS AND REMOTE SENSING: DJEBEL BOUTALEB (NORD -EST OF ALGERIA) H. GHARBI, S. CHERMAT, M.GANA
21	APPLICATION OF A FOOD SAFETY SYSTEM (HACCP) TO IMPRUVE THE QUALITY OF TAPIOCA YOGURT PRODUCT V. M. MOGA, M. A. TIŢA
30	TRADITIONAL USES, PHYTOCHIMICAL AND BIOLOGICAL ACTIVITIES OF PULICARIA ARABICA (L.) CASS. A. SASSOUI, D. SARRI, N. HENDEL, M. SARRI
36	CIRCULAR ECONOMY APPROACH IN FISHERY AND AQUACULTURE. ROMANIAN STUDY CASE L. GACEU
44	ETHNOBOTANICAL STUDY AND INVENTORY OF MEDICINAL PLANTS IN HAMMAM DALAA (M'SILA, ALGERIA N. DJERMANE, K. REBBAS, S. BENHISSEN, A. BELKASSAM
56	PHYTOCHEMICAL STUDY OF MEDICINAL PLANT «THAPSIA GARGANICA» F. MERATATE, A. LALAOUI, K. REBBAS, G. BENAICHE, H. BENSEDDIK, H. MERATATE, S. AKKAL, L. DJARRI, I. DEMIRATAS
62	PRELIMINARY RESEARCH ON CONSUMER ACCEPTANCE OF 6% REPLACEMENT DEGREE PROTEIN FISH POWDER IN SNACKS O.B. OPREA, L. GACEU
67	EFFECT OF THE INCORPORATION SPIRULINA ON THE PHYSICOCHEMICAL PARAMETERS OF A GRAPE JUICE D. DEFFAIRI, A. AMEL, D. ALILI, F. BOUGHERRA, Z. ABDELLAOUI, N. HADJADJ, N. SERIER BOUCHENAK, M. ABDI, R. FEDDAD, A. DOUMANDJI, K. REBBAS, S. AKKAL
76	ETHNOMEDICINAL USES, PHYTOCHEMISTRY AND BIOLOGICAL ACTIVITIES OF TALGHOUDA (BUNIUM FONTANESII BATT. AND RELATED SYNONYMS): A REVIEW N. ADOUI, H. BENDIF, A. BENABDALLAH, N. SOUILAH, N. DAOUD, M.D. MIARA

PROPEERTIES OF WOOD BIOMASS FROM COMPOSITE MATERIALS

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Abstract: Renewable energy in 2020 accounted for 67% of total primary energy consumption in the European Union. Countries with a high production of renewable energy are Germany, Spain, Denmark, France. Renewable energy sources are the energy of the sun, wind, water, biomass.

Most of the production of renewable energy belongs to biomass, about 47% of the total energy produced by renewable energy sources, followed by hidro energy 45%.

The energy potential of renewable sources has different intensity, for example solar energy is different in certain months of the year. In the hot summer months the sun emits an amount of energy of 1.49 kWh/ m^2 /day and in the cold months we have an estimate 0.34 kWh/ m^2 /day.

Keywords: calorimetric bomb, biomass, calorific value, composite materials

1. Introduction

Biomass is a primary source of carbon along with other renewable energy sources. It can be used as a raw material for the production of energy, biofuels and biochemical fuels, in order to obtain energy independently of the region or country. The lower calorific value of wood varies between 15480 and 19440 kJ/kg (4300 and 5400 kWh/t), depending on the species and moisture content. Biomass can have a potential of 14% of the world's total energy needs.

Low bulk density, high moisture content, degradation during storage and low energy density of lignocellulosic biomass, pore a number of significant challenges in the field of scientific research.

Storing a large amount of wet biomass will increase costs through the high rate of dry matter loss. Thermochemical conversion technologies (pyrolysis, gasification, heat processing) together with the co-generation of biomass in coal fired power plants are important in the production of thermal energy.

There are 10 million installations in China, which means that in rural areas these installations provide over 80% of the necessary production.

In Germany, Austria, Sweden, there has been a dynamic in recent years of sales of stoves and

boilers that use as fuel in the form of pellets and other solid biofuels.

In Germany, in 2020, the purchase of biomass thermal power plants was 11000 pieces. European Union directives seak to achieve energy saving standards by limiting the amount of biodegradable wate stored.

Carbon dioxide emissions from burning one kg of biomass replace the use of 0.33 m³ of methane gas, 0.74 kg of coal, 0.43 kg of fuel oil. Approximately 280 TWh of bioenergy, representing 1.5% of global energy in 2010 and 9 EJ of bioenergy, was used in the industrial sector. The cost of electricity from renewable sources is 270 EURO/MWh, and for mixed combustion biomass 120 EURO/MWh.

The estimated annual amount of wood biomass that could be produced by the introduction of energy crops would be 7200-32000 tons/year for a cultivated area of 2100 ha. The amount of renewable energy that could be obtained would be 216-960 GWh/year.

Traditional biomass (wood, coal, agricultural waste) is used for cooking and water heating. The heat generated by biomass can be a cost-competitive option. The amount of carbon stored in the biomass should be about 50% of the weight of the biomass, and through the heat treatment processes the amount of carbon increases and decreases the mineral substances.

The processing and drying of the material is established according to the European standards CEN/TS 14961 and STAS 9279-89.

According to the standards, the optimum moisture content for storing minced meat is 30%, and for use in thermal power plants it shouldn't exceed 20% moisture content.

The physical characteristics as well as the chemical composition of the biomass act on the quality of the wood material as a solid fuel. The characteristics of biomass differ from one species used as fuel to another. The moisture content can vary between 20-55 % compared to the dry mass of the firewood material, or at moisture content lower than 10% for pellets and briquettes. Final qualities can be improved by wood pretreatment technologies.

The traditional use of wood is generally low in efficiency (10%) and produces significant emissions (dust, noxious gases, fine ash).

Tehnological development has led to the implementation of highly improved pellet heating systems. Their efficiency compared to fireplaces is better. Fireplaces may have a negative efficiency during the year. Internal heating is advanced and can achieve efficiencies of 70% - 90% with low emission. These systems are found in Scandinavia, Austria and Germany. In Sweden, high performance heating systems have been developed with energy biomass pellets, which are burned in automated combustion systems.

2. Materials and method

The process of determining the caloric value of the wood materials refers first of all to the preparation of the raw material and the installation, than to the actual determination and finally to the obtaining of the final result. The preparation of the wood material for testing consists in taking a small part of about 0.6-0.8 grams of the whole material, the sample weighed with an accuracy of 0.0002 g.

The sample must be clean, made of freshly cut wood, because old wood doesn't have all the volatile and flammable substances that could influence its calorific value. This sample is placed in a porcelain crucible and placed in a laboratory oven for drying at a temperature of $103 \pm 2^{\circ}$ C.

The preparation of the installation for the test refers to the verification of the amount of water in the calorimeter (such as to exceed by 1-2 mm the cover of the calorimeter pump), of the

agitator A of the water Ap in the tank, of the computer software C, the thermometer outside the calorimeter T and the gas pressure level in the oxygen cylinder B₀. Test sample 1 is connected to the cotton thread 2 and placed in the pump crucible 3. Connect the spiral nickel wire 4 of the sample and the cotton thread, after which the protective cover 5 is correctly positioned. The crucible is connected to the calorimeter pump cover 6 by two electrodes 7 and 8, which continue with the electric coupling wires of the calorimeter pump 9 and 10. By screwing the pump cover, the pump 11 is connected through the nozzle 12 to the oxygen cylinder B_0 , introducing 30 atmospheres. Insert the pump into the calorimeter of the Cu installation, connect the two electronical wires, close the calorimeter cover and the thermostat T to determine the temperature (Fig.1).

Enter the computer software, complecty the type of test (determination or calibration), the name of sample the mass of the sample, the nickel and cotton thread, as well as other necessary data.

Next, the operation of determining the caloric power starts, selecting and activating the "STARTS" button on the computer program displayed on the monitor (Fig. 2). From this moment on the actual test to determine the caloric power begins.

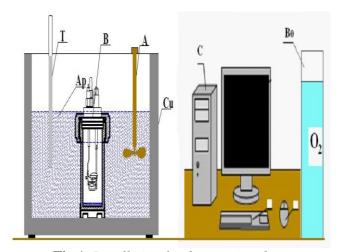


Fig.1. Installation for determining the calorific value of wood biomass with calorimetric pump with software

The test contains three different periods, respectively:

- The initial period (fore), which aims to determine the temperature variations, of the water in the calorimeter, due to the exchange of heat with the outside before burning. During this thermocuple temperature is displayed and read period, usually 5 minutes, the precision minute by minute.

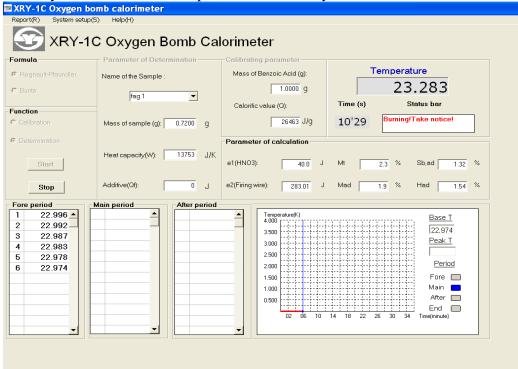


Fig. 2. Description of the process to assess the caloric power

- The main period ("main") begins with the ignition of the sample and has as a consequence the increase of the water temperature in the calorimetric vessel, due to the burning of the wood particle and the emonation of heat.
- The final period ("after") aims to determine the average temperature variation of the water in the calorimeter due to the heat exchange with the outside, after combustion

For the OSB (Oriented strand boards) with thickness 18 mm, at U= 0%, m= 0.5560 g, higher caloric power= 19346 kJ/kg, lower caloric power= 18751 kJ/kg, density= 0.505 g/cm³, U= 10%, m= 0.5940 g, higher caloric power= 17130 kJ/kg, lower caloric power= 16810 kJ/kg, density= 0.540 g/cm³, U= 20%, m= 0.7930 g/cm³, higher caloric power= 15213 kJ/kg, lower caloric power= 14572 kJ/kg, density= 0.720 g/cm³, U=50%, m= 0.6520 g, higher caloric power= 9460 kJ/kg, lower caloric power= 7858 kJ/kg, density= 0.592 g/cm³.

The variation of the calorific power for OSB is presented in Fig. 3.

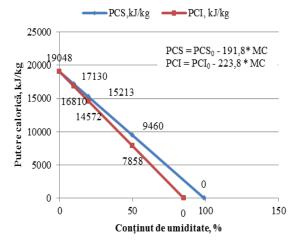


Fig. 3. The variation of the calorific power for OSB

For the densified laminated wood, at U= 0%, m= 0.8000 g, higher caloric power= 19483 kJ/kg, lower caloric power= 19004 kJ/kg, density= 0.727 g/cm³, U= 10%, m= 0.6440 g, higher caloric power= 17056 kJ/kg, lower caloric power= 16736 kJ/kg, density= 0.585 g/cm³, U= 20%, m= 0.8320 g/cm³, higher caloric power= 14870 kJ/kg, lower caloric power= 14230 kJ/kg, density= 0.756 g/cm³, U=50%, m= 0.7700 g, higher caloric power= 8310 kJ/kg, lower caloric power= 6710 kJ/kg, density= 0.700 g/cm³.

The variation of the calorific power for densified laminated wood is presented in Fig. 4.

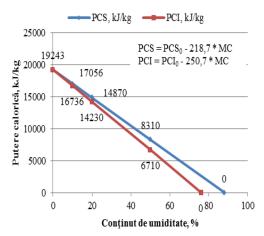


Fig. 4. The variation of the calorific power for densified laminated wood

3. Conclusions

Combustion is one of the most important thermo-chemical processes for producing energy. Combustion processes a possible only in the presence of oxygen, which is usually introduced into the hearth through the combustion air. The great potential in energy production is held by biomass (47%) followed by hydro energy (45%), at European level.

Increasing the calorific value of biomass through dry processes is a current direction of research and investment in all countries of the world. Current research shows that biomass roasting leads to increased caloric density.

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PLANT COVER ANALYSIS AND DIACHRONIC EVOLUTION USING GIS AND REMOTE SENSING: DJEBEL BOUTALEB (NORD -EST OF ALGERIA)

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Abstract: The forest vegetation of Djebel Boutaleb is experiencing degradation in its entirety under the effect of burly anthropogenic pressure and climatic risks. Hence, the vegetation cover requires analyzes to conserve and protect it with a better knowledge of the composition and the vegetation dynamics of this ecosystem.

For that, spatio-temporal studies of vegetation evolution have been applied using multitemporal Landsat imagery. An NDVI classification technique was used to generate thematic maps, with 4 major classes. The use of remote sensing data and geographic information system (GIS) has made it possible to detect important changes in vegetation cover over 35 years.

The results showed that between 1984 and 2019, the overall estimation of the evolution of the vegetation of Djebel Boutaleb is positive; with a negligible strong gain estimated 0.17% and an insignificant progression estimated at 72.67%.

Keywords: Djebel Boutaleb, Evolution, GIS, NDVI, Remote Sensing, Vegetation

1. Introduction

Algeria's northern regions, where climatic and environmental conditions help the development of forest formations, cover 250,000 km², i.e. a little more than 10% of the total area. (Ghennai, 2014) [1].

Algerian forests have experienced, over time, numerous aggressions by man through his different activities which threaten their sustainability; either intentionally and by need, ignoring their impacts on the environment; or involuntarily persuaded that the resources they offer are infinite [2].

In Djebel Boutaleb, which is part of the Mediterranean forests, the forest vegetation, under the effect of anthropogenic actions, worsened by climatic conditions is experiencing degradation in its entirety [3-7]. Hence, the vegetation cover requires analyzes to conserve and protect it with a better knowledge of the

composition and the vegetation dynamics of this ecosystem. Studies which are interested in its vegetation are less important than that of geological ones.

The Boutaleb Massif has not been subject to any diachronic study. This is the reason we proposed, for the first time, to study its vegetation cover regression with a spatiotemporal study using Geographic Information System (GIS) and remote sensing.

Among the works carried out on cartography:

- Chalifoux et al, (2006);
- Alhaskeer (2013);
- Chevigny (2014);
- Molina et al, (2016) [8-11];
- Gana (2017a);
- Gana (2017b);
- Gana (2018) [12, 13, 14];
- Benkaci et *al.* (2018);
- Arfa (2019);

- Gharbi and Chermat (2021);
- Meliani (2021) [15-18].

The aim of this study is to show the importance of using remote sensing in knowing the state and the spatio-temporal evolution of vegetation between 1984 and 2019. The data obtained in this work will be organized in a geographical database that will supply specific information for the management and protection of natural resources in general and particularly floristic.

2. Materials and Methods

2.1. Studied Area

Boutaleb forest massif is located in the southern part of Setif, between $35^{\circ}~41'~30''N$ latitude and Est $5^{\circ}~13'~30''E$ longitude. It situated

70 km from Setif and extends from west to east over 35 km ,with an estimated extension of 15 km along a horizontal plane (as the crow flies) and an area of almost 33,000 Ha.

Its altitudes are between 705 m and 1875 m (fig.1), occupy 44.24% (between 1076 and 1275 m) and 38.28% (between 1276 and 1375 m).

The highest point is Djebel Afghane peak, which reaches 1875 m in the central part of the massif. Elsewhere, the peaks are also relatively high, Soubella 1548 m, Bouhellal 1837m, Chehellou 1781m, Bouiche 1717m, and Aouzourt 1672 m. It is almost wholly from the Lower Cretaceous, characterized by marly and dolomitic soils (Ghennai, 2014).

Most of its surface is covered by the Upper Aptian, Albian and the Barremian Aptian lands (Lower Cretaceous and Upper Cretaceous) (forest conservation) [19].

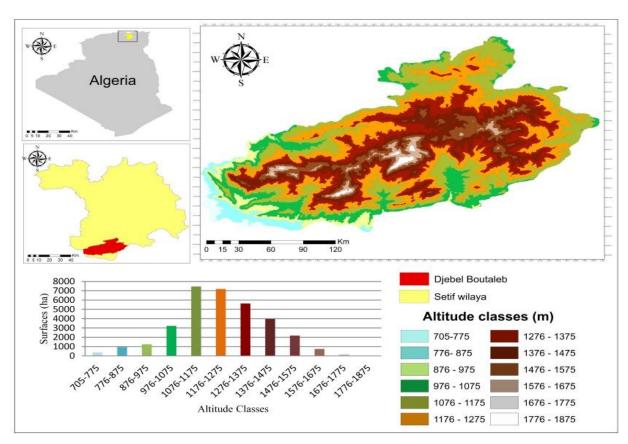


Fig.1. Geographic location and altitude classes of the study area

The reliefs are in their entirety extremely rough. The main slope categories are 10-25° and 25-45° with respectively 33.4% and 35.4% of the total area.

The slopes are oriented from north to south with an average of 5,755,936 hectares for northern exposure and 17.07% for southern exposure. The rate of other exposures varies from 0.43 to 13.4% (fig. 2). The hydrographic network

of this forest massif is very intense, but lacking a permanent superficial hydrography, nearly all the rivers that are spread in the massif are temporary, dry in summer, with the exception of Oued Faragh whose permanent flow joins Oued Soubella to the west, the latter flows into the chott of Hodna (sedjar, 2012).

Emberger's quotient Q2 analysis shows that it is between 29 and 66, and the minimum temperature of the coldest month is between -3.6 and -0.6°C, this allows the accomplishment of the bioclimatic floors map.

The latter shows that Jebel Boutaleb is characterized by 2 bioclimatic floors: arid in cold

winter in the north and south and semi-arid in cold winter in the rest of the massif.

The different vegetation floors defined in the study area are:

- Thermo-Mediterranean (<1300 m);
- Meso-mediterranean (1300 m-1500 m):
- Oro Mediterranean (1400-1800 m);
- Alti Mediterranean (> 1800 m)

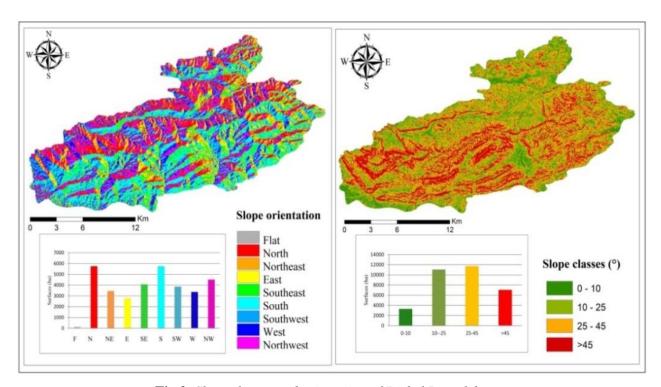


Fig.2. Slope classes and orientation of Djebel Boutaleb

The vegetation is distributed as follows:

- Tree vegetation: dominated by *Cedrusatlantica* (3%) and *Pinushalepensis* (29.53%) (table 1). While for *Quercus ilex*, it only exists in the massif in the matorral form.
- Shrub vegetation: represented by high, dense, perforated and clear matorrals made from *Quercus ilex, Cedrus atlantica* and *Pinus halepensis*.

We also find medium perforated matorrals which are formations based on *Phillyrea*

angustifolia, Pistacia terebinthus and Pistacia lentiscus and clear matorrals dominated by Juniperus phoenicia and Juniperus oxycedrus.

- Herbaceous vegetation: is well represented by scratched lawns, dominated by herbaceous chamaephytes and thorny xerophytes in pads occupying the summits of certain Djebel, steppes, which are open formations of xerophytes in tufts colonizing nearly the entire southern flank of the massif and finally, ermes therophytics which invade the forests and the matorrals clearings (forest Conservation).

Table 1. Forest stands types distribution in Djebel Boutaleb

Forest stands	Surface (ha)	Surface(%)
Juniperus L., matorral	912.75	3
Quercus ilex L., coppice	2479.78	8.15
Natural regeneration	384.09	1.26
Reforestation	1929.02	6.34
Pinus halepensis Mill., forest	8981.6	29.52
Juniperus Phoenicia L.,- Quercus ilex L., matorral	3307.35	10.87
Quercus ilex L., matorral	2890.11	9.5
Juniperus L., scrubland	1240.7	4.08
Cedrus atlantica Manetti., forest	910.92	2.99
Other	7385.81	24.28
Total	30422.13	100

2.2. Data Sources

The data used for this study include: data collected from fieldwork and Landsat satellite data (thematic mapper 5 and operational land imager 8) was used, which are available from United States Geological Survey (USGS) website http://earthexplorer.usgs.gov/ [20].

For the choice of the optimal date of the image, it is essential to obtain an image without

cloud and without overlapping and distinguish all the elements of the land while avoiding confusion between the vegetation floors [21].

We opted for the period from June to August (table 2).

The ancillary data used in this study includes: The Arc GIS (10.1) and ENVI (5.1) software's were used for realization of change maps, cartographic database and spatial analyses.

Table 2. Characteristics of the Landsat images used

	Tubic It enter the Edition of the Editional interger th				
Characteristics	16/07/1984	18/08/1996	03/08/2008	19/08/2019	
ID	LT05_L1TP_194	LT05_L1TP_194	LT05_L1TP_194	LC08_L1TP_194	
	035_19840716_2	035_19960818_2	035_20080803_2	035_20190802_2	
	0171213_01_T1	0180215_01_T1	0160116_01_T1	0190819_01_T1	
Format	TIFF	TIFF	TIFF	TIFF	
Data	Landsat 4-5 TM	Landsat 4-5 TM	Landsat 4-5 TM	Landsat 8 OLI	
Image centre	194/35	194/35	194/35	194/35	
Acquisition date	16/07/1984	18/08/1996	03/08/2008	02/08/2019	
Spatial Scale (m)	30m	30m	30m	30m	

2.3. Methodology

We have used Landsat satellite images and Google Earth images, whose combination allowed us to produce more accurate maps (Meliani et *al*, 2021).

The satellite images were processed after delimitating the study area and eliminating all parts of the image outside the area [22].

The "raw" image produced by the sensor cannot be used directly because it has errors linked to the sensitivity of the sensors, the geometry of observation, and the presence of the atmosphere's effect around the earth.

Lighting conditions and atmospheric attenuation are highly variable in time and space. So, the images need atmospheric corrections in order to homogenize the multispectral data.

The principle of this correction is to convert digital image counts (gray level) into luminance at the satellite sensor level and then remove the disturbing effects of the atmosphere from this new variable in order to eventually result in a physical measurement of the reflection at the target surface [23]. We used the ENVI classic tool (quick atmospheric correction).

Among all the vegetation indicators proposed since the origins of spatial remote sensing, the normalized vegetation indicator has

become the standard tool for describing the spectral behavior of vegetation cover. This indicator, most often called NDVI according to its English abbreviation, is calculated from two spectral bands, infrared IR (where the vegetation cover has high reflectance) and red R (where mineral surfaces have high reflectance) according to the following expression: NDVI = (IR - R)/(IR + R).

The resulting neo-channel shows an increasing gradient of plant activity ranging from black (absence of covering) to white (very high chlorophyll activity).

The result of an NDVI takes the form of a new image, the value of each pixel being between 0 (bare soil) and 1 (maximum vegetation cover). It is the analysis of the range of shades extending between these extreme values (very rare) that informs the observer about the density of the plant cover and the quantity of green biomass [24].

The next step consists in discovering the major changes between the different dates chosen. This allows detecting the slightest pixel changes between two images at different dates (1984-1996, 1996-2008 and 2008-2019). The images obtained show, both spatially and quantitatively, the important changes that occurred in vegetation over time in the study area.

Possible changes can be: strong progression, little progression, stabilization, strong regression

and little regression. The last step is to make maps of vegetation change over 35 years.

3. Results

3.1. NDVI Results

We examined the distribution of vegetation cover over the whole study area at each date. NDVI values were variable, the highest NDVI values are found in areas with elevation varying between 1276 and 1875m, indicating a high density of vegetation cover.

NDVI values from (0.40 - 0,20) were found in areas with 876 to 1275m elevation and indicating areas with moderate vegetation cover. Low NDVI values (< 0.20) are located at regions with 705 to 875m elevation indicates croplands and no vegetated areas: built-up, water, rocks... (fig.3).

NDVI differentiation data layers were generated for each image pair to identify the different pixel changes.

The modified zones are seen in dark green, light green, red and pink, represent the strong progression, the insignificant progression (almost stability), the strong regression and the insignificant regression (almost stability) of the vegetation zones respectively, while the regions without change are displayed in white.

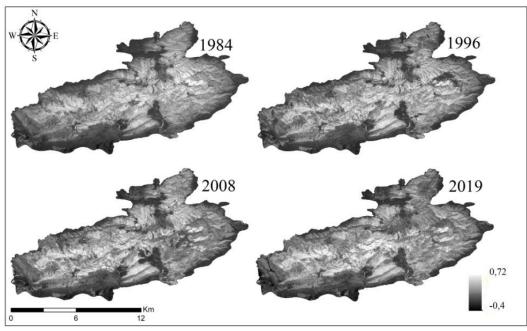


Fig. 3. NDVI map of Djebel Boutaleb in 1984, 1996, 2008 and 2019

3.2. Change Detection Analysis

The results obtained through the analysis of NDVI, illustrate a substantial change in vegetation between 1984-1996, 1996-2008 and 2008-2019 (fig 6, 7 and 8)

3.2.1. Global analysis of the evolution of the vegetation between 1984 and 2019

Map 04 shows the important changes that have occurred in the Boutaleb massif over a period of 35 years. The vegetation of Djebel Boutaleb has experienced a strong increase of 55,17 ha in the center, the North, the North-West and the South-West and a sharp regression of

0,75 ha in the western part. During the same period, the areas of insignificant progression of vegetation are in the majority, they occupy an area of 86.23%, while the area of the zones where the regression is insignificant is estimated at 13.56%.

The evaluation of the evolution of the vegetation, over the whole period, remains minor with a strong progression of only 54.72 ha i.e. 0.17% of the massif total area, while for the insignificant progression; it is estimated at 72.67%.

For a detailed follow-up, we have subdivided the study period into 3 periods: 1984-1996, 1996-2008 and 2008-2019.

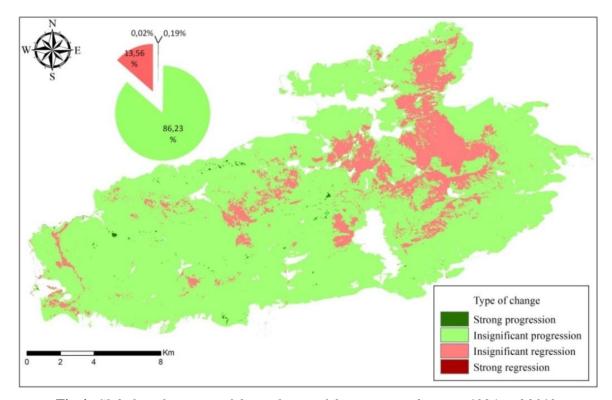


Fig.4. Global analysis map of the evolution of the vegetation between 1984 and 2019

3.2.2. Analysis of the evolution of the vegetation between 1984 and 1996

Figure 5 shows that the vegetation cover has seen an important progression by 431.28 hectares i.e. 1.3% particularly in the West and the South. Against this gain, 2.34 ha or 0.007% of the strong

regression was recorded in the east and northeast. We also notice a progression and an insignificant regression estimated at 94.34% and 4.35% respectively.

The massif then shows a positive balance with a strong regression of 428.94 ha and an insignificant regression of 91.3%.

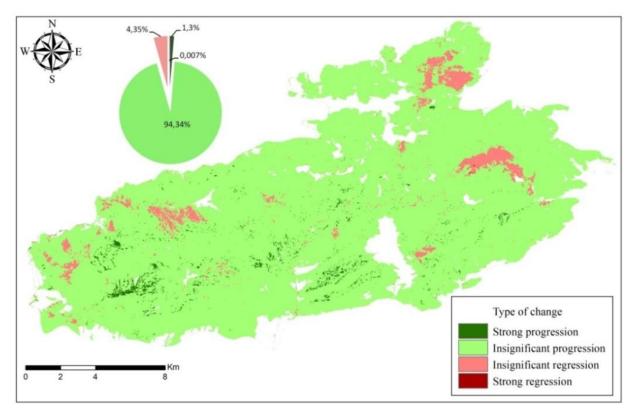


Fig.5. Analysis map of the evolution of the vegetation between 1984 and 1996

3.2.3. analysis of the evolution of the vegetation between 1996 and 2008

According to figure 6, we notice that the areas of strong progression are only 0.01%. The strong regression affected 4.73%, it is located in the eastern part with some markings in the north, west and southwest. We also noted that the areas

where no significant regression was recorded are the majority, they occupy 92.16%. On the other hand, the tiny progression occupies 3.1%.

Djebel Boutaleb recorded then a negative balance with a strong regression for the surface area equivalent to 1560.96 hectares and 31 049.19 ha for a low significant regression.

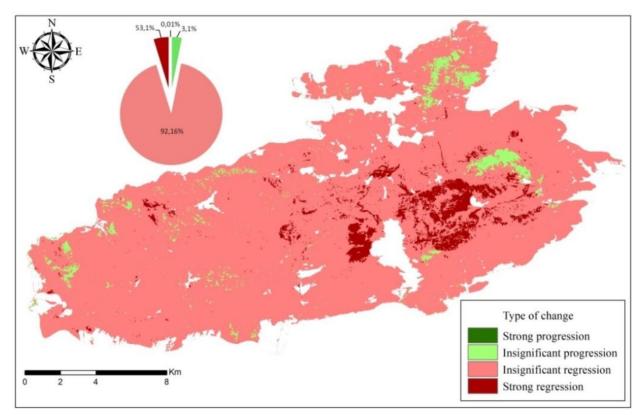


Fig.6. Analysis map of the evolution of the vegetation between 1996 and 2008

3.2.4. analysis of the evolution of the vegetation between 2008 and 2019

From figure 7, we can see that between 2008 and 2019, 91.54% of the vegetation cover in Djebel Boutaleb experienced little progression. At the same time, 8.2% registered a slight regression.

The strong progression in the center and northwest is about 6 times greater than the strong regression in the west.

It is estimated at 72.81%. So the massif shows a positive balance with a strong progression of 0.18% and a slight progression of 83.34%.

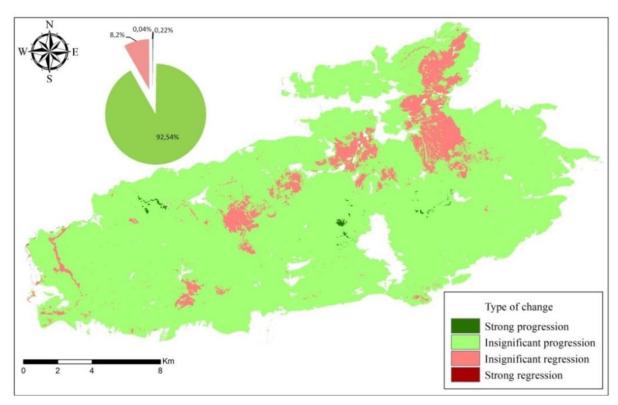


Fig.7. Analysis map of the evolution of the vegetation between 2008 and 2019

4. Discussion

The analysis of the three maps allows us to note that the greatest progression relates to the period 1984-1996.

This progression is mainly due to natural regeneration and also to reforestation. Furthermore, the strongest regression relates to the period 1996-2008.

According to Forest Conservation Department of Setif, the main causes of this regression are much more linked to fires, where there is a loss of 742 ha, particularly in summer, in the state forests of aleppo pine, edar and juniper oxyhedron in the municipalities of Ainazel, Rasfa, Hamma, Boutaleb and Salah-Bey. We also have the very noticed presence of the pine processionary of the aleppo pine on the one hand, and parasitic foci on the cedars (table 3).

In addition, there is the polypore on Aleppo pine and cedar.

We can also add the growing needs in wood for various uses, forest rangelands to meet the food needs of livestock, land clearing, over grazing, mismanagement, deforestation (fig.8). However, this kind of activities could endanger the natural vegetation and reduce the forest ability to regenerate.

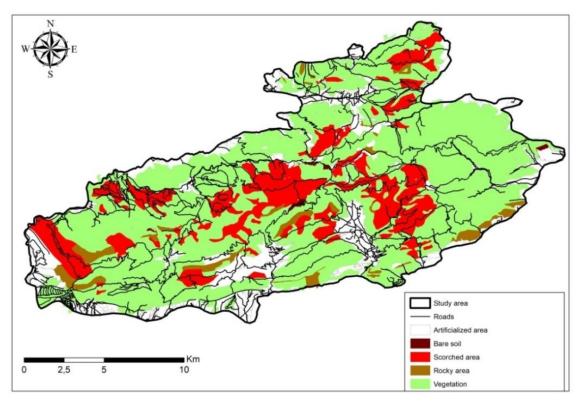


Fig.9. Land use map of Djebel Boutaleb

 Table 3. Scorched areas of Djebel Boutaleb (1985-2014)

	<u> </u>
Year	Surface
1985	5ha
1986	298ha
1987	25ha
1998	180ha
1999	155ha
2001	175ha
2002	17ha
2005	10ha
2006	5ha
2007	50ha
2008	150ha
2009	112ha
2009	12ha
2010	75ha
2012	185ha
2013	432ha
2014	167ha
Total	2053ha

5. Conclusion

In this study, we suggested to detect the change in the vegetation cover of Djebel Boutaleb over a long period using satellite images of the LANDSAT TM5 and OLI8 type by the use of the vegetation index by normalized difference NDVI to realize the change maps on each date.

Geographic information systems (GIS) together with remote sensing appear to be a tool for monitoring evolution in vegetation cover.

This diachronic study illustrates that between 1984 and 2019, the overall estimation of the evolution of the vegetation is positive; with a negligible strong gain estimated 0.17% and an insignificant progression estimated at 24,058.71 ha.

The conservation of the vegetation then requires avoiding all anthropogenic factors and particularly the repeated fires which lead to the deterioration of the vegetation.

Finally, it would be interesting to use satellite images at higher resolutions for a more detailed analysis in order to better distinguish areas of degradation or regeneration.

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APPLICATION OF A FOOD SAFETY SYSTEM (HACCP) TO IMPRUVE THE QUALITY OF TAPIOCA YOGURT PRODUCT

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Abstract: This study presents aspects of the implementation of the HACCP system in order to identify potential hazards and assess hazard analysis and establish measures to prevent and control them, as well as to determine critical control points and establish critical control limits to ensure food safety in the manufacture of tapioca yogurt.

Keywords: HACCP, food safety, tapioca yogurt, hazard points.

1. Introduction

Acidic dairy products are among the top preferences of dairy consumers. Although studies show that the recipe for making sour milk was accidentally discovered centuries ago by nomads who noticed that under certain microbiological conditions and at a certain temperature the milk coagulates, it has not been changed much. over time [1].

The preservation of milk by acidification is still practiced today, the range of acidified dairy products has increased (sour cream, sana, yogurt, kefir, etc.) with the discovery of the coagulation potential that different and varied microorganisms have on milk.

Interventions on the manufacturing process included the addition of various ingredients in order to strengthen the dairy product, improving the nutritional value and sensory characteristics, but also the physico-chemical characteristics.

Yogurt is perhaps the best known acidic dairy product obtained by fermenting milk under the action of two microorganisms: Lactobacillus bulgaricus and Streptococcus theromophilus.

A common defect in yogurt is the removal of whey, called syneresis.

The appearance of this defect negatively influences the consumer's perception of the product. To eliminate these defects, a number of additives are widely used in production systems in order to improve the quality of yogurt: powdered milk derivatives, powdered vegetable derivatives, starch, gelatine, hydrocolloids and others [2-12].

The addition of tapioca powder to yogurt has beneficial effects on the texture and consistency of yogurt, implicitly on consumer perception. A market study conducted in Sibiu County shows that there is interest from yogurt consumers for our product, even if some consumers have not tried tapioca products. This may be due to curiosity and high interest in exotic products and a healthy lifestyle [13].

The nutritional value of tapioca powder in yogurt is minimal, due to the chemical composition of tapioca powder. Using 2% tapioca powder in the manufacture of yogurt, better physico-chemical results were obtained in terms of acidity, lactose content and syneresis, compared to the classic yogurt, without additives and the yeast with 2% added skimmed milk powder.

Regarding the sensory characteristics, following the comparative analysis between the 3 yogurt samples, we found that the yogurt with tapioca has a better consistency and is more homogeneous [14].

The production of safe food for consumption should be a key objective for food companies. HACCP - Hazard Analysis. Critical Points Control is a preventive control system for food quality assurance. Applying the principles of this program helps to prevent, reduce and even eliminate the risks that may arise throughout the production process [15].

In the European Union, the implementation of HACCP is mandatory for all small and medium-sized food companies.

2. Materials and methods

Starting from the technological scheme of the tapioca yogurt product, the aim of the research is to lay the foundations of a HACCP plan following its principles and specific steps:

- product description
- defining the use of the data and identifying the category of consumers of the product
- technological flow chart

- assessment of the hazard analysis and establishment of prevention or control measures
- determination of critical control points (CCP) for the identified hazards
- establishing the critical limits to be observed for each CCP identified

3. Results and discussion

A. Product description

Table 1. Product description

YOGURT WITH TAPIOCA	
Product name	Yogurt with tapioca
Ingredients	Pasteurized cow milk, tapioca, lactic cultures for yogurt
Organoleptic characteristics	Colour: Uniform Milk-specific white
	Taste and Smell: specific to yogurt -pleasantly sour. Foreign taste
	or smell is not accepted.
	Texture: homogeneous
Physio-chemical characteristics	Fat 3.5 ±1%
	Total solids content min. 11%
	Acidity min. 0.6% lactic acid
	Protein substances min. 2.8%
Microbiological characteristics	Salmonella, Enterobacter, E. coli, Shigella, Klebsiella - absent
Packing method	In 150 g glass jars and the closure with metal lids.
Storage instructions	Refrigerated rooms, clean, disinfected, ventilated, no foreign
	smell at temperatures between 2–8 °C.
Terms of validity	21 days*

B. Identification of the category of consumers of the product

Tapioca yoghurt addresses, first of all, the market segment of informed consumers, who have difficulties in finding on the Romanian market a traditionally prepared yoghurt, little processed, without additives, which exceeds the quality of yoghurts on the market.

Tapioca yoghurt is addressed, secondly, to consumers of natural yoghurt, without the addition of fruits, cereals, cocoa, with a fat content of 3.5%. Compared to this product segment, tapioca yoghurt has a firmer texture, a better bound curd, does not contain protein additives from milk, milk powder or modified starch [13].

C. Flow Diagram

The flow diagram presented in the fig. 1. shows all the stages of the technological process, from the reception of the raw material to the sales level. After constructing this diagram, the food safety team checks on the spot, after which they approve the flow chart.

D. Hazard Identification and Determination

Identifying potential risks is an important step in implementing the HACCP system. Once potential hazards have been identified, their analysis or control measures are established.

For the manufacturing process of tapioca yogurt, the following table (table 2) ilustrates the identification of hazards, their causes and the appropriate prevention measures. The identification of hazards has been classified according to their nature:

- F: physical
- C: chemical
- B: biological

Control measures are those actions and activities that can be used to prevent risks, to eliminate them or to reduce their impact or the likelihood of their occurrence at an acceptable level.

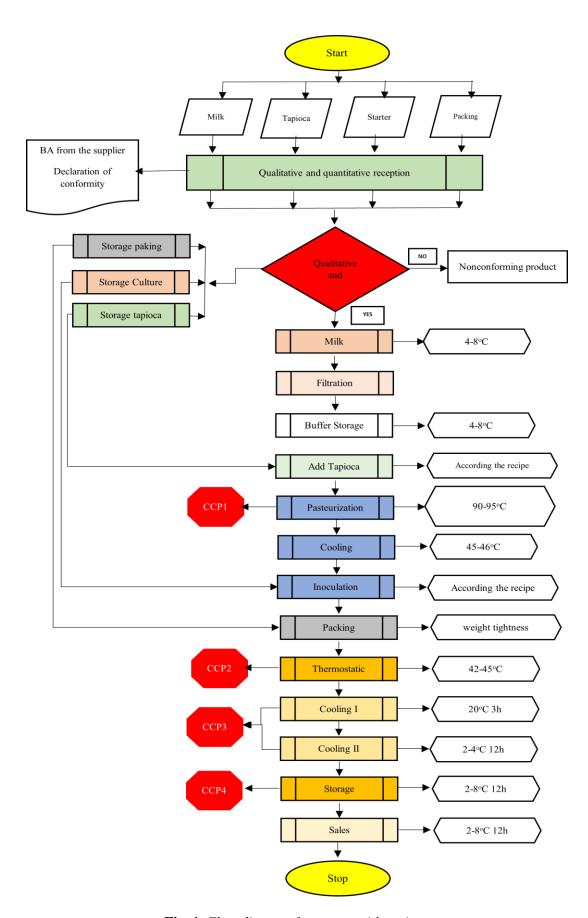


Fig. 1. Flow diagram for yogurt with tapioca

Table 2. Hazards Identification

			T	Table 2. Hazards Identification			
Nr	Process stage	Potential hazards	Is the	Cause	Preventive		
•		physical, chemical,	danger		measures		
		biological	potentiallly				
			signigicant?				
1.	Quantitative	B: contamination with	Yes	- Non-compliance	-return of the raw		
	and	microorganisms		with sanitary	material to the		
	qualitative	(Mycobacterium		norms at	supplier		
	reception of	tuberculosis,		producers	-supplier audit		
	raw matereal	Salmonella, E coli,		1	TT		
		Staphylococcus					
		aureus, Brucella					
		campylobacter,					
		Listeria					
		monocytogenes,					
		Bacillus cereus,					
		Mycobacterium bovis)					
		C: Pesticides,	yes	- animal feed is			
		Antibiotics, drugs,		contaminated with			
		detergents etc.		agricultural			
		C		fertilizer			
				- drug-treated			
				animals			
		F: Presence of foreign	no		- Milk filtration		
		bodies: hair, metallic					
		impurities					
2.	Cleaning	F: the filter surface is	no		- Maintenance		
		perforated			filters		
		B: microorganisms on	yes		- Checking the		
		the filter			hygiene and		
					operation of		
					the filter		
3.	Cooling and	B: abnormal growth	yes	-Failure to	- Periodic		
	buffer storage	of microorganisms		observe the	temperature check		
				temperature			
				regime			
4.	Addition of	F: erori de dozare	no	- Staff inattention	- Staff training		
	tapioca			- Calculation			
		.		errors	_		
5.	Pasteurization	B: incomplete	yes	- Failure to	- Temperature		
		destruction of		comply with	check		
		microorganisms		pasteurization	- Performing		
				parameters	physico-chemical		
					analyzes to verify		
		Contomination		immoner	pasteurization		
		Contamination wirh		- improper	- sanitizing the		
		detergents		hygiene of the machine	machine		
6.	Cooling			macinne			
7.	Insemination	C: Contamination	-	immen	- agniticina 41		
/.	msemmation		no	- improper	- sanitizing the machine		
		with detergents		hygiene of the machine	macinie		
		F: Foreign bodies:	VAS	maciille	Staff training		
		r. Poteigh bodies:	yes		- Staff training		

		hair, metallic impurities from the staff			
8.	Packing	B: contaminare cu microorganisme	-	- Improper sanitation of production spaces and equipment - Improper hygiene of staff	 Improving hygiene conditions Periodic checking of the health of the staff
		C: Contamination with detergents	no	- improper hygiene of the machine	- sanitizing the machine
9.	Thermostatic	B: improper development of milk acidifying bacteria Development of other microorganisms	yes	- Improper temperature - Non-compliance with the hygiene conditions from the previous steps	- Observance of the thermostating regime and periodic temperature verification - Observance of hygiene conditions throughout the technological flow
10.	Cooling	B: increasing acidity by continuing the development of microorganisms	no	- Failure to observe the cooling temperature	Observe the cooling regime I and II and check the temperature regularly
11.	Storage	B: development of unwanted microflora	yes	- Improper temperature and relative humidity conditions in the storage room	Periodic checking of temperature and humidity Sanitation of storage spaces periodically

E. Hazard assessment

Based on the cause-effect diagram (ISHIKAWA), the severity of the hazard and its probability of occurrence were determined. These values (between 1 and 3) are set by the designated team based on their experience.

The following table (table 3) shows the hazard analysis according to the level of probability of occurrence of a hazard and its severity, by multiplying them. [16]

F. Identification of Control Points (CP) and Critical Control Points (CCP)

Once the potential hazards that can be encountered in the production of tapioca yogurt have been identified, critical control points and control points are determined.

A decision tree was used to perform this analysis as you can see in table 4.

Those risks that obtained a rating ≥ 3 were passed through this decision tree. [15]

G. Identification of critical limits

values that allow control to be ensured. the critical limits are set out in Table 4.

Once the critical control points have been established, it is necessary to establish the limit

 Table 3. Hazard assessment

F	Table 3. Hazard assessi					
Nr.	Process stage	Potential hazards physical,	Severity	Probability	Hazard	
		chemical, microbiological			rating	
1.	Quantitative and	B: contamination with	3	1	3	
	qualitative reception	microorganisms (Mycobacterium				
	of raw matereal	tuberculosis, Salmonella, E coli,				
		Staphylococcus aureus, Brucella				
		campylobacter, Listeria				
		monocytogenes, Bacillus cereus,				
		Mycobacterium bovis)				
		C: Pesticides, Antibiotics, drugs,	2	1	2	
		detergents etc.				
		F: Presence of foreign bodies: hair,	1	2	2	
		metallic impurities				
2.	Cleaning	F: the filter surface is perforated	2	1	2	
		B: microorganisms on the filter	3	1	3	
3.	Cooling and buffer	B: abnormal growth of	3	1	3	
	storage	microorganisms				
4.	Addition of tapioca	F: erori de dozare	1	1	1	
5.	Pasteurization	B: incomplete destruction of	3	1	3	
		microorganisms				
		Contamination wirh detergents	2	1	2	
6.	Cooling	-				
7.	Insemination	C: Contamination with detergents	2	1	2	
		F: Foreign bodies: hair, metallic	2	1	2	
		impurities from the staff				
8.	Paking	B: contaminare cu microorganisme	3	1	3	
		C: Contamination with detergents	2	1	2	
9.	Thermostatic	B: improper development of milk	3	1	3	
		acidifying bacteria				
		Development of other				
		microorganisms				
10.	Cooling	B: increasing acidity by continuing	3	1	3	
		the development of microorganisms				
11.	Storage	B: development of unwanted	3	1	3	
		microflora				

Table 4. *Identification of CCP and CP*

Process stage	Potential	N	Nr.question (fi	rom the decision to	ee)	CCP/
	hazards physical, chemical , biochimi cal	Q1 Are control measures in place for the hazard?	Q2 Does the process step eliminate or reduce the hazard to an acceptable level? NO – Q3	Q3 Could contamination with the hazard occur at unacceptable level(s) or increase to unacceptable level(s)? NO is not CCP	Q4 Will a subsequent process step eliminate or reduce the hazard to an acceptable level?	СР
		not CCP YES – Q2	YES – is CCP	YES – Q4	CCP YES – is not CCP	
Quantitative and qualitative reception of raw matereal	В	Yes	No	No	-	СР
Cleaning	В	Yes	No	Yes	Yes	CP
Cooling and buffer storage	В	Yes	No	Yes	Yes	СР
Pasteurization	В	Yes	Yes	-	-	CCP 1
Packing	В	Yes	No	No	-	CP
Thermostatic	В	Yes	No	Yes	No	CCP 2
Cooling	В	Yes	No	Yes	No	CCP 3
Storage	В	Yes	No	Yes	No	CCP 4

Table 5. Critical limits for CCP

Stage	ССР	Critical limits	Ways of	Corrective actions
			surveillance	
Pasteurization	CCP1	Time: 20 min Temperature 90-95°C	Check the temperature every 5 minutes	If the value to is less than the accepted value, the automatic valve must close the access to the tank and the pasteurization must be repeated. If the temperature is lower, the pasteurization time increases
Thermostatic	CCP2	Temperature 42-45°C Time: 2.30 h pH: 4.6-4.7 Total adidity: 80- 90°C	Periodic temperature check every 10 minutes Performing periodic physico-chemical analyzes (pH and acidity) after 1.30h and 2h and 2.30h	Staff training Periodic maintenance of equipment
Cooling	CCP3	I: Temperature 20°C Time: 3 h II: Temperature 2-4°C Time: 12 h	Periodic temperature check once an hour	Periodic maintenance of equipment
Storage	CCP4	Temperature: 4-6°C Recomanded: 10 days	Periodic temperature check once an hour	Periodic maintenance of equipment

Once the critical limits have been set, monitoring procedures will be established. Persons responsible for monitoring critical limits will be appointed to ensure continuous monitoring of production.

Another important step is to establish a corrective action plan that includes a corrective action plan that will be applied when the critical limits have been exceeded (table5).

4. Conclusions

This study is intended to demonstrate once again the applicability, functionality and effectiveness of the HACCP system.

The application of the HACCP system will certainly have a positive impact by implementing in reducing or even eliminating the risks when making tapioca yogurt.

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TRADITIONAL USES, PHYTOCHIMICAL AND BIOLOGICAL ACTIVITIES OF *PULICARIA ARABICA* (L.) CASS.

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Abstract: This article aims to overview the traditional uses, chemical compositions and biological activities of essential oils and extracts of Pulicaria arabica which is considered a traditional medicinal plant in the treatment of various diseases.

The P. arabica is used in the treatment of several diseases such as antidiarrheal, anti schistosomiasis, digestive disorders, ulcers, anti-tobacco and skin diseases. Several different biological properties have been reported such as antimicrobial, antioxidant, analgesic, antipyretic, anti-inflammatory, insecticidal, hepatoprotective and nephritic effects. The chemical investigation of the P. arabica (aerial parts, leaves, stems and flowers) showed some qualitative and quantitative differences in the chemical composition, for example the major compounds for essential oils (Thymol, γ -Cadinene, α -Cadinol and epi- α -Cadinol), flavonoids and Caryophyllene derivatives.

Keywords: Asteraceae, *Pulicaria arabica* (L.) Cass., Ethnomedecine, Chemical composition, Bioactivities.

1. Introduction

Pulicaria arabica (L.) Cass. (Asteraceae, Inuleae) its basionym are Inula arabica L. This species its native range is South and East Mediterranean basin to Pakistan and Arabian Peninsula (Fig.1a) [1]. There are 12 species of Pulicaria that grow naturally in Algeria [2,3], among them P. arabica is a grass that shelters wet places and marshes. The plant is traditionally used in Algeria, Egypt, Saudi Arabia and other countries [4-6].

The phytochemical study of the essential oil of *P. arabica* showed a significant chemical variation in different countries such as Egypt [7-10], Saudi Arabia [11-13], Algeria [4,14].

Some publications reported bioactivities of *P. arabica* as antibacterial [4,15-17], antioxidant [4,16], antiviral [18], analgesic, antipyritic and anti-inflammatory in hepatic and nephretic [19] and insecticidal [14].

This article aims to give the relevant literature of the medicinal uses, chemical compositions, and biological activities of *P. arabica* growing in Algeria comparing with other countries.

2. Taxonomic tree of *Pulicaria arabica* (L.) Cass.

Kingdom : Plantae
Subkingdom : Tracheobionta
Order : Asterales
Division : Magnoliophyta
Family : Asteraceae
Class : Magnoliopsida
Subclass : Asteridae

Genus : *Pulicaria* Gaertn.

Species : Pulicaria arabica (L.) Cass.

3. Vernacular names of *Pulicaria arabica* (L.) Cass.

Arabic: Rara ejub, abu- ain-safra, deithouth,

hatassa, arba'a Aiuyûb.

English: Ladies' false fleabane French: Pulicaire d'Arabie

Spanish: Arrudena, coniza, yerba de las pulgas,

yerba pulguera

Portugal: Herba piolheira, tagueda.

4. Botanical description

Perennial plant with yellow flowers (Fig.1b) [1], erect 10-60 cm, with erect branches. Dichotomous, paniculate inflorescence. Capitula heterogamous, multi-florous, long stalked, large or fairly large, 10-20 mm in diameter, with spreading ligules, fairly long, amply exceeding the involucre and flowers of the disk the tubular and hermaphroditic.

Receptacle flat, bare, with alveolate spines. Involucre (bracts on a few rows, generally narrow, the outer ones are short or foliaceous) densely hairy, hispid achenes with fairly long double pappus, the outer one is short, free, cupuliform, made up of flakes more or less horny at the base, free or more or less fused with the inner, the inner is composed of long, scabrous bristles.

Leaves are linear, oblong lanceolate, entire, slightly or subdentate, not embarrassing, nor rolled up, tuberculous, rough, hairy or not [2].

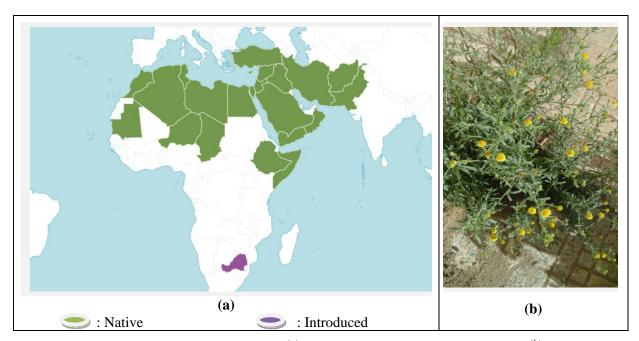


Fig.1. Distribution in the world^(a) [1] and specimen harvested in Algeria^(b) of P. arabica (L.) Cass.

5. Traditional uses

Pulicaria species are used in the treatment of several diseases such as hypoglycemia, fever, spasmodic diseases, cancers, microbial and

inflammation [20,21]. However, the *P. arabica* (L.) Cass. is used in folk medicine in the form of various remedies (Table 1).

Table 1. Traditional uses of *P. arabica* (L.) Cass.

Traditional uses	References
- antidiarrheal agent and antischistosomiasis	[6]
- treatment of painful swellings and boils	[4]
- treatment of digestive disorders	[5,12]
- treatment of ulcers	[22]
- anti-tobacco	[18]
- use as lotion for curing skin diseases	[23]
- the powder of the plant is used as an sneeze, by nasal intake, in the treatment of certain Oto-Rhino-Laryngology diseases and headaches	[24]

6. Phytochemical investigation

The analysis of chemical components identified in Pulicaria genus essential oils shows that the oil consists of several groups of components, which are monoterpènes hydrocarbons, oxygenated monoterpenes, sesquiterpene hydrocarbons, oxygenated sesquiterpenes, aldehydes, and ketones [21].

In addition, Table 1 summarizes the previous investigations of authors on the phytochemical studies from *P. arabica* that characterize the presence of volatile oil [4,11,14,25], flavonoids [7,10,12], and caryophyllene derivatives [8]. The

comparison of the chemical composition of the essential oil of *P. arabica* (aerial parts, leaves, stems and flowers) shows some qualitative and quantitative differences in the composition of the oil from Saudian, Tunisian and Algerian of the plant studied [4,11,14,25].

Other research on the *P. arabica* species has reported the isolation of certain flavonoids compounds and caryophyllene derivatives (Table 1).

Table 2: Major chemical components of *P. arabica*

Table 2: Major chemical components of F.			
Country	Parts / Major components	References	
-	A) Essential oils		
Algeria	Aerial parts ■ Bicyclo(4.4.0) dec-1-ene2-Isopropyl-5-methyl-9-methylene (17.2%) ■ 1H-indene.1-ethylideneoctahydro (13.2%) ■ Δ-Cadinene (13.0%) ■ α-Muurolene (5.9%) ■ β-Ocimene (5.8%)	[4]	
	Aerial parts ■ epi-α-Cadinol (23.9%) ■ δ-Cadinene (21.1%) ■ α-Cadinol (19.8%) ■ Germacrene D-4-ol (8.4%)	[14]	
Saudi Arabia	Aerial parts ■ β-Selinen-2α-ol (3.4%) ■ β-Cedrene (2.5%) ■ Carbonyl compounds and acids (2%) ■ (Z)-Jasmone	[11]	
Tunisia	Stems ■ Thymol (15.2%) ■ cis-Calamenene (9.1%) ■ Carbonyl compounds and acids (7.5%) ■ α-epi-Cadinol (6.6%) Flowers ■ γ-Cadinene (11.7%) ■ cis-Calamenene (9.5%) ■ α-Cadinol (8.6%) Leaves ■ α-Cadinol (10.3%), ■ α-epi-Cadinol (8%) ■ cis-Calamenene (7.9%)	[25]	
	B) Flavonoids		
Saudi Arabia	Aerial parts ■ Quercetagetin-3,6,7-trimethyl ether	[12]	
Egypt	Leaves and flowers ■ Quercetagetin-3,7-dimethyl ether ■ Quercetagetin-3,5,7-trimethyl ether	[7]	

■ Quercetagetin-3,5,7,3'-tetramethyl ether ■ Quercetin 3-glucoside	
■ 3-glucuronide	
Aerial parts	
■ Quercetagetin-3,5,6,7,4'	[10]
■ 3,5,6,7,3'-pentamethyl ethers	r - J
■ Quercetagetin-3',4'-dimethyl ether	

Table 2: Major chemical components of *P. arabica* (Continued)

Country	Parts / Major components	References
	C) Caryophyllene derivatives	
Egypt	Aerial parts	
	■ 12-Hydroxy-5β-methoxy-6(14)-dehydro-5,6-dihydrocaryophyllen-7-one	
	■ 12-Acetoxy-5β-methoxy-6(14)-dehydro-5,6-dihydrocaryophyllen-7-one	
	■ 12-Acetoxy-5α-methoxy-6(14)-dehydro-5,6-dihydrocaryophyllen-7-one	[8]
	■ 5\alpha,12-dihydroxy-5,6-dihydrocaryophyllen-7-one	
	■ 12-Acetoxy-7\alpha,14-dihydroxy-5E-caryophyllene	
	■ 12-Acetoxy-14-methoxy-5E-caryophyllen-7-one	
	■ <i>Bis</i> -[SZ-7-oxo-caryophyllene]-l4-O-ether	

7. Biological activities

In the literature, various biological activities have been reported on the genus *Pulicaria*. Most studies have been reported on antimicrobial [4,17,26-30], and antioxidant [4,27-32].

In addition, *Pulicaria* species have other activities such, analgesic, antipyretic, anti-inflammatory, hepatoprotective and nephritic effects [19], antihistaminic [33], anticancer [30,34-37], leishmanicidal [38,39] and insecticidal [14,40].

Few works have been cited on biological activities, either on essential oils or extracts of *P. arabica*. In summary, the ethanolic extract of *P. arabica* induces potent analgesic, antipyretic and anti-inflammatory with significant hepatic and nephritic protective actions.

In addition, the disturbance of liver and kidney function parameters induced by CCl₄ intoxication was recovered by *P. arabica* extract [19].

The microbiological effect of the essential oil of *P. arabica* show a significant inhibitory action against bacteria and yeasts [14], in addition, the ethanolic extract of *P. arabica* show activity against *Enterococcus faecalis*, Methicillinresistant *Staphylococcus aureus* [17];

The potential extended spectrum β -lactamase (ES β L) suppressing activities of crude extracts of P. a arabica show a inhibitory activities [15]; and that in vitro antioxidant activity of essential oils of P. a rabica was achieved using DPPH indicated a average capacity [4], but, the evaluation of the antiradical test of the methanolic extract of P. a rabica shows a high capacity [4].

Also, the essential oils of *P. arabica* show a very marked insecticidal activity against *Spodoptera littoralis* [14].

8. Conclusion

Pulicaria arabica (L.) Cass. is an important medicinal plant utilized for the treatment of different diseases. The literature revealed that the phytoconstituents and biological activities are available in the *Pulicaria* genus.

The species *P. arabica* remains the least studied species in terms of biological activities compared to other species of the genus; this article has summarized the few works cited in the literature, and our objective is to encourage researchers to value as an example the biological activities such as those of insecticidal, anti-inflammatory, anticancer and leishmanicidal of the species *P. arabica*.

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CIRCULAR ECONOMY APPROACH IN FISHERY AND AQUACULTURE. ROMANIAN STUDY CASE

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Abstract: The Blue Bio-economy and its problematic related to the capitalization of own waste represents a special chapter in the general framework of the circular economy. The reasons for the specific approaches are closely related to the complexity of the technologies used, the high nutritional value of the waste, but also the food safety issues that arise. For this reason, the elements of legislation approved at European level are highlighted and below are presented a series of statistical data that position Romania in the evolutionary framework of other European countries, from several points of view: fish production, consumer preferences, import / export balance, etc. The conclusions of the study show the need to implement technologies for capitalizing on rest raw materials (RRMs) in the fish industry.

Keywords: circular economy, fishery, aquaculture, Romanian study case

1. Introduction

The Blue Bio-economy Strategy is part of EU's strategy to increase the use of marine resources to make novel foods and additives, animal feeds, and nutraceuticals, but also pharmaceuticals and cosmetics. Enhanced food production from marine biological resources should contribute to reduced pressure on land resources for agriculture, mitigate climate change, and enable future food security and sustainability. There are several nutritional and health benefits from marine food due to the high level of proteins, omega-3 fatty acids and vitamins [1, 2, 12].

However, there is a significant potential for improvements in the marine food supply systems, since fish and other seafood constitute the commodity group with the second largest food losses and waste at 35% [3-5].

An improvement of the supply systems, e.g., by waste reduction and full utilization of the biomasses, will therefore be a valuable contribution to advancing the blue bio-economy towards increased sustainability and competitiveness.

Rest raw materials (RRMs) are the leftover materials in the preparation of the main product and may be both edible and inedible.

In general, all leftover materials from slaughtering and processing can be used in food production, either for humans, feed for food-producing animals, or for technical purposes. Currently, utilization of RRMs is typically used to create products such as fish oil, fishmeal and biogas.

Many scientific reports the pre-processing and handling methods of RRM in industry to identify available amount and potential for production of food-grade fish meal and food ingredients [12].

The technologies of enzymatic hydrolise of protein are described and are used as a reference to define the potential for increased value creation in the fishing and aqua culture industry. In addition, the market for harvested seaweed is reviewed in order to map available raw material and further establish the potential for increased product value.

Current legislation and barriers are addressing the accompanying challenges and should be essential for establishing new products on the market.

In this context, it is very important to investigate the Romanian potential of higher valorisation of fishery and aquaculture waste.

This topic is studied in the frame of the project SuMaFood (SUSTAINABLE PRESERVATION OF MARINE BIOMASSES FOR AN ENHANCED FOOD VALUE CHAIN).

2. Materials and methods

EU legislation regarding food, hygiene regulations and novel foods and ingredients legislations were studied.

Starting from EUROSTAT, EUROFISH, EUROBARAMETERS and FAO database, the main parameters regarding fishery production, fleet, aquaculture were taked into account. Purchasing factors and consumer preferences were also studied, as indicators of the potential for waste valorisation.

3. Legislation

Production of fish products intended for human consumption must comply with the Food and Hygiene Regulations set by the European Commission (EC). Relevant regulations are EC 852/2004, EC 854/2004, EC 853/2004 as amended by EC 1020/2008, and EC 2074/2005 as amended by EC 1022/2008.

The regulations provide a guide to the categorization of different rest raw material of animal origin, how they should be handled, and what it may be used for. RRM that fall into Category 1 is considered of the lowest quality, as this material may transmit diseases to humans or other animals. This material is to be kept far away from the food chain and is sent for incineration.

However, in the fishing and aquaculture industry this category is non existing. Category 2 materials encompass mostly fish with signs of clinical disease which have been treated with medications or dead fish from aquaculture. RRMs (whole fish) in Category 2 are utilized for technical purposes such as bio-energy and fertilizers. It may also be used for food for animals with the purpose of producing fur [3-5].

Rest raw material arising at a fish slaughterhouse, fish landing facility or fish

processing plant which is handled in compliance with legislation of hygienic processing, is termed Category 3 material and is allowed to be sold for human consumption. Material not handled according to the legislation of hygienic handling but the animal byproduct legislation, is termed byproduct category 3 material and may be utilized for foddering livestock [5] dead fish where the cause of death is not related to infections or disease, but from lack of oxygen or invasion of jellyfish or algae.

In order to ensure the safety of the consumers, regulation must be in place to supervise the entire value chain from harvest, transport, processing and all the way to the end product. The producers must confirm that the raw materials are captured or harvested in a sustainable way and by certified operators, and it must be assured that the material is processed in approved facilities and in accordance with laws and regulations.

The producers must also keep information on how the raw material has been handled and whether it complies with the Food and Handling regulations set by the EU commission. If the raw material is to be part of a new product to be placed on the market, additional requirements also must be met.

The authorization and use of novel foods and food ingredients have been harmonized in the European Union since 1997 when Regulation EC 258/1997 on novel foods and novel food ingredients was adopted [6].

If it cannot be documented that the raw material was utilized as food within the European Economic Area (EEA) prior to 15th of May 1997, an approval must be given by the EU commission before the food can be placed on the market. EU legislation and EFSA guidance documents detail how to compile dossiers for submission and the information and studies required for the evaluation and approval [6].

4. Results and discussion

Romania is a minor EU producer of fishery products. As for aquaculture production, in 2019 it ranked 19th among EU-28 (i.e. UK included) producing countries (tables 1, 2) [7-11].

Table 1. Romania	ı in the	world	and	in the	EU

(1.000 tones)	World	EU - 28	Romania	% world	% EU - 28
Catches	93.519	4.824	11	0,012%	0,23%
Aquaculture	120.104	1.367	13	0,011%	0,94%
Total	213.623	6.191	24	0,011%	0,38%

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Table 2. Fishing fleet

Vessels (2019)		Capacit	y (2019)	Power (2019)		
Number: 167		GT:	1.582	KW: 6.547		
Total FTE: 59,75 (2018, source: JRC)						
0 - 11m		12 - 23 m	24 – 39 m	> 40 m		
Jobs (FTE): 46%	Jobs	s (FTE): 40%	Jobs (FTE): 13	3% Jobs (FTE): 0%		

^{*} GT: Gross tonnage; KW: Kilowatt; FTE: Full Time Equivalent

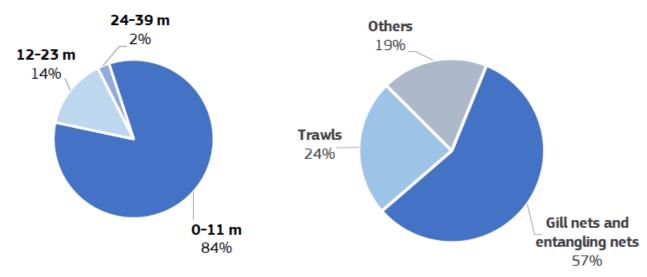


Fig. 1. The Romanian fishing fleet by length and gear (% of total no. of vessels)

LANDINGS (Source: EUMOFA, based on Eurostat data)

Landings comprise the initial unloading of any fisheries products from on board a fishing vessel in a given Member State. They include aquatic plants and species not destined for human consumption. Landings are recorded in net weight and value, and concern landings made by vessels from EU Member States, Iceland, Norway and the UK.

In 2019, landings in Romania only include fresh products destined for human consumption (figure 1 - 4).

There are 2 fishing ports registered in Romania (source: EU Master Data Register, 26 November 2021).

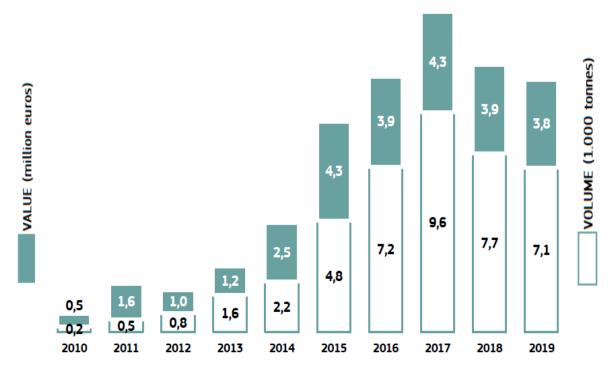


Fig. 2. Total landings. Values are deflated by using the GDP deflator (base=2015)

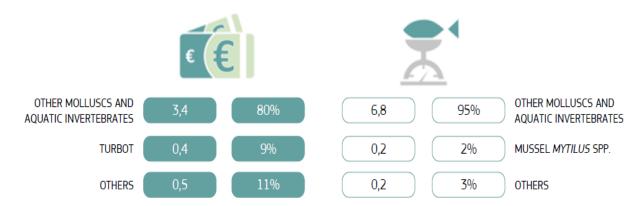


Fig. 3. Main commercial species landed and % of total (2019, million euros (nominal value) and 1000 tonnes)

AQUACULTURE (Source: EUMOFA, based on Eurostat and FAO data)

Aquaculture consists in the farming of aquatic (fresh or saltwater) organisms, such as fish, molluscs, crustaceans and aquatic plants. Aquaculture data are reported in live weight equivalent and value.

According Eurostat, the aquaculture activity in Romania is limited to freshwater farming. The following main production methods were used in 2018:

- 79% of production: in ponds;
- 21% of production: in tanks and raceways;

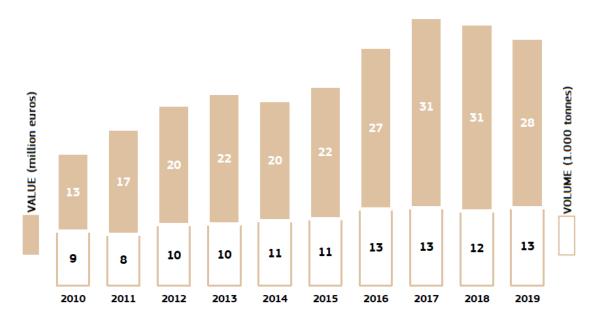


Fig. 4. Total production. Values are deflated by using the GDP deflator (base=2015)

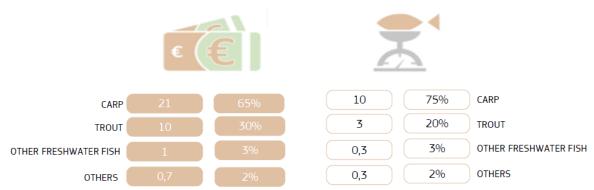


Fig. 5. Main commercial species farmed and % of total (2019, million euros (nominal value) and 1000 tonnes)

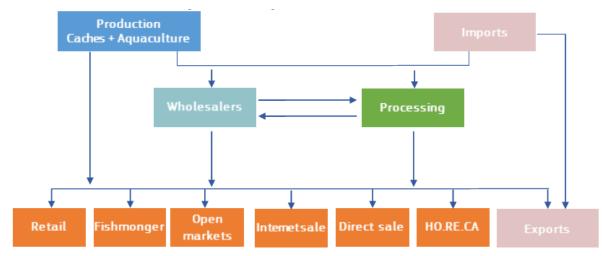
PRODUCER ORGANISATIONS (Data as of 15-12-2021, source: DG MARE)

In Romania, 11 producer organisations (POs) are formally recognized. Their role is to contribute to the achievement of the objectives of the Common Fisheries Policy (CFP) and of the Common organisation of the markets (CMO) through the collective management of their members' activities. Of the 11 POs, 9 operate in the fishery sector and 2 in aquaculture. (fig. 5, 6)

PROCESSING

According to Eurostat-SBS, 1.303 persons were employed in the Romanian fish processing industry in 2019.

The sector recorded a value added of 11,5 million euros, covering 1% of the value added of total manufacture of food products. In 2020, the main products sold were "Prepared or preserved crustaceans. molluscs and other invertebrates (excl. chilled, frozen, dried, salted or in brine, crustaceans, in shell, cooked by steaming or boiling) (excl. prepared meals and dishes)" and "Prepared or preserved fish (excl. whole or in pieces and prepared meals and dishes)", "Frozen whole salt water fish" (source: Eurostat-PRODCOM). (fig.



Source: Eurofish

Fig. 6. The supply chain of fisheries and aquaculture products in Romania (source: Eurofish)



Fig. 7. Consumer preferences on purchasing channels (source: Eurobarometer, 2021)

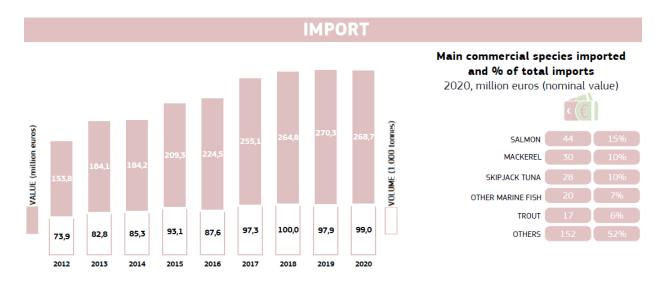


Fig. 8. Main commercial species imported and % of total imports

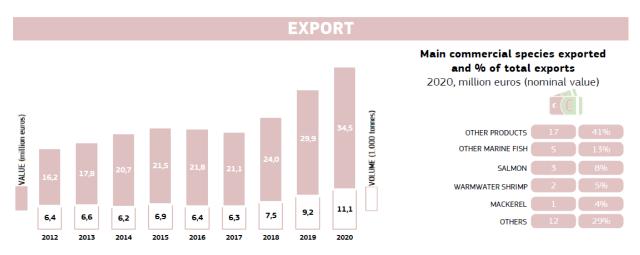


Fig. 9. Main commercial species exported and % of total exports

CONSUMPTION

In 2019, apparent consumption was estimated at 8,11 kg per capita, a 1% increase compared with 2018. The most consumed species were mackerel, carp and herring (source: EUMOFA). Regular consumers, namely those who eat fishery and aquaculture products at least once a month, mainly belong to age groups 40-54 and 25-39. Young people (15-24) are less inclined to

consume fish in Romania, as well as at EU-28 level. In this category, regular consumers cover 59% of the total, which is lower than at EU level (67%, UK included). Romanians consume especially fresh and frozen products (fig. 10); loose fish (63%) is less frequently consumed than at EU level (68%, UK included) (source: EUMOFA, "EU consumer habits regarding fishery and aquaculture products", 2017).

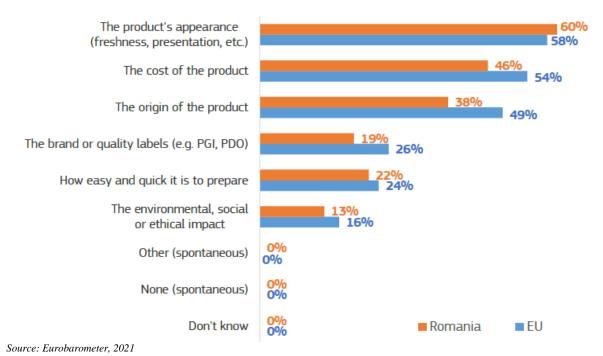


Fig. 10. Purchasing factors (8,11 kg per capita in live weight equivalent, 2019, source: Eumofa)



Fig. 11. Preferences regarding wild or farmed product (source: Eurobarometer, 2021)

Conclusions

The results of the latest research in the field of food security show that in order to ensure the food of the next generations, it is necessary to capitalize on the superior waste of the food industry.

In this context, the fish processing industry has a special potential by transforming its own waste into protein powders (by enzymatic hydrolysis), respectively calcium powders (obtained by crushing the bones free of animal protein).

Romania currently has a modest portfolio in terms of fishery and aquaculture production, but market demand is growing. Therefore, concerns for the recovery of this waste are important, with many local producers showing their willingness to use new recovery technologies.

Acknowledgement

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ETHNOBOTANICAL STUDY AND INVENTORY OF MEDICINAL PLANTS IN HAMMAM DALAA (M'SILA, ALGERIA)

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Abstract: The ethno-botanical surveys carried out using questionnaires in the region of Hammam Dalaa made it possible to inventory 68 medicinal species belonging to 35 botanical families and to collect as much information as possible concerning local traditional therapeutic uses. This study also showed that the leaves are the most used part. Recipes are basically prepared by decoction or powder. Of all the diseases treated, digestive disorders are the most frequent. These results are considered as a source of information for scientific research in the field of phytochemistry and pharmacology.

Keywords: M'Sila, Hammam Dalaa, medicinal plants, ethnobotany.

1. Introduction

Medicinal plants continue to meet an important need, despite the existence and influence of modern health systems, about 35,000 species of plants are used worldwide for medicinal purposes, which forms the largest range of biodiversity used by humans [1].

These medicinal plants are important for pharmacological research and drug synthesis not only when their constituents are used directly as therapeutic agent but also as raw material for drug synthesis or model for pharmacologically active compounds [2].

Today, herbal treatments are coming back to the fore, as the effectiveness of drugs such as antibiotics (which is considered the near universal solution to serious infections) decreases as bacteria and viruses are adapted to drugs and their are increasingly resisting [3].

The most serious diseases, cancer, sclerosis which are treated in a very difficult way, but thanks to Phytotherapy which is an important

alternative can bring comfort in the classic treatment of these serious diseases [4].

Medicinal plants constitute a precious heritage and a real treasure for humanity, and are in great demand in the world and more particularly in developing countries [5].

These medicinal plants are still a source of medical care in developing countries due to the lack of a modern medicinal system [6].

Traditional herbal medicine was well developed in Algeria, but the use of conventional medicine is the cause of a neglect of these ancestral practices, which risk falling into oblivion [7].

Alongside the programs of some international organizations such as the World Health Union (IUCN), which is interested in promoting the conservation of biodiversity and the sustainable use of natural resources in North Africa, and also the involvement of local communities in the biodiversity conservation [6].

The carrying out of ethnobotanical surveys in the region of Hammam Dalaa aims to obtain a floristic inventory of the medicinal plants used by the population and the collection of as much information as possible on the therapeutic uses practiced in the study area.

The preservation of this knowledge is a challenge for the conservation and development of resources and this within the framework of the sustainable development of the study area.

2. Materials and method

2.1. Geographical location of the study area

The study area was located north of M'Sila. This area is limited to the north by El M'hir, to the east by Maadid, to the west by Ouanougha and to the south by the commune of M'Sila (Figure 1).



Fig. 1. Geographical location of the study area

2.2. Natural conditions

The region of Hammam Dalaa does not have any weather station. The closest posts are those of M'sila and Bordj Bou Arreridj. The average annual rainfall is 221 mm at the M'Sila station; on the other hand, they are 385 mm at the Bordj Bou Arreridj station.

The mountain ranges receive larger quantities of water, of the order of 400–500 mm in the Saharan Atlas and possibly reaching more than 600 mm in the Hodna and Aurès-Belezma mountains [8].

Hammam Dalaa have a semi-arid climate characterized by cold, harsh winters and hot, dry summers.

2.3. Ethnobotanical surveys

The ethno-botanical surveys on medicinal plants were carried out during the months of March, April and May 2020 using a questionnaire sheet, these surveys allowed us to draw up a list of the medicinal plants used by the population of Hammam Dalaa in traditional herbal medicine.

There were three methods of ethno-botanical surveys, which were the most used, and the most appropriate to our case study, where we completed 132 survey leaves with informants who have knowledge of the therapeutic use of plants.

a. Rural population surveys

This survey consists of asking questions to the villagers about the plants used in traditional medicine, the parts of the plant used, the methods of preparation, and the types of diseases treated by each plant.

b. Surveys of herbalists

The survey of herbalists makes it possible to draw up a list of spontaneous medicinal plants in the study area. This investigation which carried out during the purchase of the plants, enabled us to collect necessary information concerning the vernacular medicinal plants exposed to the sale, the therapeutic uses, the posology and the diseases treated by each plant.

c. Surveys of traditional healers

The survey showed that some family members have significant knowledge of plants of medicinal interest and possessing healing gifts. The pharmacological knowledge of plants from traditional healers makes it possible to identify the basic concepts of the perception of the natural environment and the description of diseases [9].

2.4. Questionnaire leaves

The tool of our survey was a form consists of two parts. The first was based on the person surveyed (age, sex, level of education and professional situation), the second part collected information about each plant medicinal plant studied, this information makes it possible to assess the knowledge of the plant, its use, the prescription and the method of preparation recommended by each of the people interviewed.

3. Results and discussion

The results obtained from the ethno-botanical surveys were expressed in technical leaves, which consist in highlighting the characteristics of the medicinal plants used by the population of the study area. Gives us a list of medicinal plants used by the local population of this area and their therapeutic properties and traditional uses (Table 1, Annex).

3.1. Choice between traditional medicine and clinical medicine

The ethno-botanical survey of medicinal plants carried out with the population of the study area, and the collection of data concerning therapeutic uses, allowed us to describe, classify, and inventory medicinal plants, this floristic inventory highlights a floristic richness of 68 species belonging to 35 botanical families.

Regarding the uses of medicinal plants, and the treatment based on these plants. Show that the population uses phyto-therapy only or with clinical medicine with a cumulative percentage of 60% and the rest 40% uses clinical medicine, which was explained that the population interested by traditional therapies to treat their ailments, and by the fact of the effectiveness of the therapeutic practices that people had acquired from their ancestors.

3.2. Use of medicinal plants according to the survey profile

Both men and women were concerned with the use of medicinal plants, however, women used traditional medicine much more than men, because women have multiple functions and responsibilities as mothers that they must take care of their families, especially their children.

Elderly people in the age group 51 to 70 used medicinal plants more than other age groups, as their knowledge and experience on the use of plants in traditional medicine.

The transmission of knowledge of the uses and properties of medicinal plants from one generation to another is in danger because it is not always ensured.

According to the censuses carried out with the people, who used medicinal plants and according to their level of study, the results obtained show that the illiterate were the most users of the medicinal species followed respectively by the people who have the secondary level, the average level then the university level.

3.3. Information about the herbal drug according to the diseases treated

Analysis of the results obtained shows that plants such as: white horehound (*Marrubium vulgare* L), thyme (*Thymus algeriensis* Boiss. & Reut.), Germander (*Teucrium polium* L), Ivette (*Ajuga iva* L), white mugwort (*Artemisia helba*

alba Asso.) were among the plants most used by the local population in traditional medicine.

On the other hand to these plants which were in great demand and used in the study area, there were other medicinal plants which have a low use because of their toxicity such as oleander (Nerium oleander L), Thapsia (Thapsia garganica L).

The leaves were the part of the plant most used in treatment preparations, followed by the other parts of the plants.

The results obtained show that most of the plants intervene in the treatment of the affections of the digestive system with a percentage of (45%) then follow the other affections.

4. Conclusion

In this study, we identified 68 medicinal plants belonging to 35 botanical families, the most important of which was that of the Lamiaceae, the leaves were the most used parts; the infusion and the decoction were the most applied methods of preparation. Digestive disorders were the most known by the population in this area.

The variation in the use of medicinal plants was linked to the profile of the people surveyed; young people generally do not know the names or the use of most plant species, on the other hand the elderly, who have experience in therapeutic uses.

Women and men have a shared medicinal knowledge. Illiterates know information about the therapeutic uses of medicinal plants much more than other literate people. The population of these regions has known the use of plants in traditional medicine.

The multiplication of these ethno-botanical studies on a national scale will make it possible to better understanding the potential in this field, to assess the risks resulting from the use of certain toxic plants and to adopt a new management approach for the safeguarding and preservation of natural resources [10].

Picking must be done with caution because the strong pressure of picking leads to a decrease in productivity and the reduction or loss of biodiversity, this way of harvesting leads to the scarcity, and even the risk of total disappearance of certain species [11].

In the absence of cultivation, many plants are threatened with extinction. In this context, we propose the cultivation of plants in this region, which have been the subject of conclusive scientific work and which are used in human therapy in many countries, because the needs of the pharmaceutical industry in medicinal plants are multiplied [12].

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Annexes





Fig. 2. General view of a matorral landscape of Hammam Dalaa (K. Rebbas, 2019)

 Table 1. List of medicinal plants used by the population of Hammam Dalaa (M'Sila, Algeria) [9-22]

Families	Species	Parts used	Therapeutic properties	Traditional uses
Abietaceae	Pinus halepensis Mill.	Buds, leaves, resins, bark.	Expectorant, aphrodisiac, spermatogenesis.	A needle decoction is used as an antiseptic, balsamic and antirheumatic.
Anacardiaceae	Pistacia lentiscus L.	Leaves, resins, roots, bark.	Astringent, antiseptic, detergent, expectorant, hemostatic, simulant, vulnerary.	The leaves and the bark are used, in decoction or in powder, in the treatment of the intestine, diarrhea and diabetes.
Apicaceae	Coriandrum sativum L.	Leaves, fruit.	Antiseptic, antispasmodic, carminative, stomachic, stimulating.	The plant is used in infusion of fresh leaves in boiling water against digestive and gastric disorders. In external use: it acts as a healing agent.
Apicaceae	Petroselinum sativum L.	Leaves, stems.	Digestive, cordial, remedial, of the kidneys, healing, fortifying of the hair.	A decoction of the leafy stems is used against kidney stones and bladder problems.
Apicaceae	Thapsia garganica L.	Roots.	Thapsia is used against rheumatism and bronchitis.	External use: an oily maceration of the crushed roots is used as a compress against rheumatic pains.
Apocynaceae	Nerium oleander L.	Leaves.	Diuretic, antidiabetic, cardiotonic.	The local application of latex is recommended to treat cases of scabies.
Asteraceae	Antemisia herba-alba Asso	Leaves, tops, roots.	Emmenagogue, stomachic, vermifuge, antispasmodic, anti gastralgic.	The leafy stem, in decoction, is very indicated in case of intestinal worms, colds, gastric pains, urinary ailments and diabetes.
Asteraceae	Artemisia campestris L.	Tops, flowers, roots, leaves.	Vulnerary, anti-haemorrhagic, diuretic.	The plant is used in the form of infusion or powder against abdominal pain, colic and menstruation.
Asteraceae	Brocchia cinerea Screw	Leaves	Anti-inflammatory, analgesic, antiseptic, antibacterial, antipyretic.	The plant is used to treat stomach pain, fever, headache and migraine, cough and joint inflammation
Asteraceae	Chrysanthemum coronarium L.	The whole plant	Anti-inflammatory, analgesic.	The whole plant, in powder form, is used against stomach ailments.
Asteraceae	Lactuca sativa L.	Leaves.	Aperitif, cardiotonic, antitussive, pectoral, anti-ulcer.	A forehead massage is done with the infusion of the leaves against sunburn.
Asteraceae	Launaea nudicaulis L. Hook .	Leaves.	Antidiabetic, calming.	The powder of the leaves is recommended against diabetes and gastric ailments.
Brassicaceae	Brassica rapa L.	Roots, leaves.	Aperitif, bechic, anti-gout, anti-rheumatic.	A fumigation of the leaves with onion, the leafy stem of white horehound, eucalyptus leaves and cloves, and fish head is effective in cases of typhoid fever.
Chenopodiaceae	Atriplex halimus L.	Leaves, seeds.	Diuretic, emollient, laxative, emetic.	Mixing the powder of the plant with olive oil is very

				effective in treating broken bones.
Chenopodiaceae	Spinacia oleracea L.	Leaves.	Laxative, hepatic, anti-inflammatory of the urinary tract, anti-ulcer, anti-anemic.	A decoction of the leaves, against inflammations of the digestive tract, liver and bladder.
Cistaceae	Cistus albidus L.	Leaves, flowers.	Hypoglycemic, diuretic, healing.	The leaves, in decoction, are used against gastric pains and considered hypoglycemic. as a poultice: they are used against abscesses. In external use: it is used as a poultice against wounds.
Cucurbitaceae	Colocynthis vulgaris L.	Fruits.	Purgative, emetic, scalp tonic.	Very dilute infusion of pruned dried fruit or pulp as a purgative and hypoglycemic.
Cucurbitaceae	Cucumis citrullus (L) Ser.	Fruits.	Diuretic, hypotensive, softening, refreshing.	One drinks a spoon of oil extracted from the seeds every morning on an empty stomach against hypertension.
Cupressaceae	Juniperus oxcycedrus L.	Leaves, fruits, roots, wood, cones, resins.	Antiseptic, diuretic, stomachic stimulant, sudorific.	Cade oil extracted from <i>Juniperus oxcycedrus</i> cures rashes.
Cupressaceae	Juniperus phoenicea L.	Twigs, fruits, wood.	Antiparasitic, antiseptic, astringent.	The leaves are used, in decoction, as a hypoglycaemic. Infusion of the leaves, is used as a body bath to treat rheumatism.
Ericaceae	Arbutus unedo L.	Fruit leaves.	Anti-inflammatory, antiseptic, astringent, diuretic and depurative.	A decoction of the leaves is used against urinary calculi.
Euphorbiaceae	Euphorbia guyoniana Boiss. and Reut.	Stems.	Calms pain due to scorpion stings.	External use: The stem latex is applied to sites of viper bites to relieve pain and stop the spread of venom.
Fabaceae	Vicia faba L.	Seeds.	Laxative, lowers blood cholesterol levels.	A powder of broad bean seeds mixed with chickpea seed, rice seed, and egg yolk are used as a poultice on the face against abscesses.
Fabaceae	Ononis spinosa L.	Flowers, roots.	Antiseptic, astringent, depurative, diuretic, sudorific.	The infusion of the roots increases the secretion of urine and acts against gout.
Fabaceae	Calycotome spinosa (L.) Lamk.	Flowers, leaves and seeds.	The calycotome is recommended for external use, against swelling, edema and especially against urine retention because its active substances are strongly diuretic.	The infusion or decoction of the flowers and leaves of the calycotome is diuretic. It can be used in powder form to treat new wounds.
Fabaceae	Retama retam webb.	Whole plant.	Healing recommended to treat eye irritations, diarrhea, feverish illnesses and solitary worms.	Mixing the dried leaves with olive oil or water is effective against eczema.

Fabaceae	Trigonella faenum-graecum L.	Seeds.	Softening, anabolic, emollient, febrifuge, galactagogue, hypoglycemic, tonic.	Internally, a decoction of the seeds is used to soothe abdominal pain, cough and diarrhoea. It promotes weight recovery and acts as an aperitif and soothing. Externally, the seeds are used to remove dandruff from the hair.
Fagaceae	Quercus ilex L.	Bark, leaves, stems.	Antiseptic, antidiarrheal, astringent, haemostatic febrifuge.	As a gargle: 15g per liter of water, against angina, stomatitis and pharyngitis.
Globulariaceae	Globularia allypum L.	Leaves.	Astringent, laxative, cholagogue, depurative. stomachic and sudorific.	The infusion of this plant is recommended to treat gastric disorders, diarrhea and menstrual pain. In powder: it is used against eczema and burns.
Lamiaceae	Ajiga iva (L.) Schreb.	Whole plant.	Antiseptic, parasiticide, hypoglycemic.	Ivette tea is used against diabetes, hypertension, diarrhoea, gastric pain and cancer.
Lamiaceae	Marrubium vulgare L.	Flowering tops, leaves.	Bitter tonic, cholagogue, regulates heart rhythm.	Infusion of horehound leaves is used against colds, fever and cases of allergy.
Lamiaceae	Mentha pulegium L.	Leaves, flowering tops.	Hypotensive, bechic, pectoral, cephalic, antirheumatic, antipyretic.	In decoction, the dried or fresh leaves recommended to treat abdominal ailments.
Lamiaceae	Mentha spicata L.	Leaves	Analgesic, antiseptic, antispasmodic, aromatic, carminative, cholagogue, digestive, stimulant, tonic.	Among the villagers, spearmint is commonly used in infusion as carminative, odontalgic, tonic, stomachic, calming, hypotensive and hypocholesterolemic. It is used to treat dysmenorrhea, urinary incontinence and oral diseases. In powder, spearmint is used to treat wounds and burns as well as to nourish the hair.
Lamiaceae	Mentha viridis L.	Leaves, stems.	Antispasmodic, antirheumatic, antiemetic, antineuralgic.	The leafy stem, infused in tea, is used as a digestive and refreshing. As a poultice, the leaves are used against headaches and wounds.
Lamiaceae	Teucrium polium L.	Leaves, tops, flowers.	Anti-inflammatory, astringent, detergent, febrifuge, hypoglycemic, bitter tonic.	The plant used in infusion against gastrointestinal pain, fever, diarrhea, liver attacks, biliary retention.
Lamiaceae	Thymus algeriensis Boiss. & Reut.	Whole plant.	Vulnerable, vermifuge, tonic, stomachic, expectorant, bechic, carminative, appetizer, antiseptic.	This plant is used in the form of herbal tea to treat the flu.
Lamiaceae	Rosmarinus officinalis L.	The leaves and flowers.	Anti-inflammatory, antiseptic, antispasmodic, astringent, carminative, cholagogue, emmenagogue, febrifuge,	Rosemary leaves are used in the form of decoction or infusion against gastric disorders, colic and menstruation and back pain

			general stimulant, stomachic, tonic, vulnerary.	It is considered carminative and diuretic.
Lauraceae	Laurus nobilis L.	Leaves, fruit.	Stomachic, carminative.	Compress bay leaves, used against rheumatism.
Liliaceae	Allium cepa L.	Bulbs.	Treatment: infected wounds, otalgia, chest, otitis, eyes, constipation, headaches, rheumatism, apples, respiratory, hair loss.	The bulb of the onion is used in the form of poultice against dermatological affections, and orally against affections at the level of the gallbladder.
Liliaceae	Allium sativum L.	Bulbs.	Garlic is effective against corns, warts, earaches, arthritis pain and rheumatism.	It is considered hypotensive.
Liliaceae	Asphodelus microcarpus Salzm and Vivo	Tubers.	Detergent, resolving, antirheumatic, analgesic, antispasmodic.	An oily maceration of the tubers is applied against ear infections, rheumatism, dental pain.
Lythraceae	Lawsonia inermis L.	Leaves.	Anti-ulcer, fungicide, anti-diarrheal, vermifuge.	The leaves are used as an infusion against diarrhea and renal lithiasis.
Malvaceae	Malva sylvestris L.	Leaves, flowers, roots.	Mallow acts as a calming and diuretic.	The leaves and flowers in infusion are used against chronic constipation, cough and bronchitis.
Moraceae	Ficus carica L.	Fruits.	Laxative, anti-animic, bechic, pectoral, remedying warts.	In infusion, the fruits are used to treat cough. and the latex is used externally against warts.
Oleaceae	Jasminum fruticans L.	Flowers.	Calming and sedative.	Usually comes in the form of herbal tea made from the flowers. Jasmine essential oil is used on the skin of the face, for its antioxidant and anti-wrinkle effect but also for its antibacterial action for acne cases.
Oleaceae	Olea europea L.	Leaves, barks, fruits.	Leaves and bark: astringent, diuretic, febrifuge, tonic, hypotensive, hypoglycemic. oil and fruit: cholagogue, laxative, emollient, sedative.	Olive oil is used as a laxative in the case of chronic constipation.
Oleaceae	Phillyrea media L.	Barks.	Antiulcer, analgesic.	The bark, in decoction, is used in the treatment of fever.
Papaveraceae	Papaver rhoeas L.	Flowers	Antispasmodic, soothing, calming, emollient, pectoral, sedative, slightly hypnotic.	The infusion of the flowers is used as a tranquilizer, and Antitussive.
Plantaginaceae	Plantago albicans L.	Leaves, roots and seeds.	Softener, astringent, emollient, diuretic and laxative.	Combine with colocynth root to make poultices in wound care. Against diarrhea.
Poaceae	Ampelodesma mauritanica (Poir.) Hard. & Schinz	Leaves	The succulent tender base of its flower stems is often eaten in the countryside; its sweetish juice is refreshing.	This plant is used in infusion to treat kidney stones and gallbladder.

Poaceae	Hordeum vulgare L.	Seeds.	Diuretic, antitussive, anti-inflammatory and detoxifying.	Barley is used in the treatment of diseases: of the stomach, tuberculosis, colon, diarrhea, leanness jaundice. Boiled barley helps cure cough.
Poaceae	Stipa tenacissima L.	Leaves.	Alfa is used in the treatment of chronic scalp ulcers.	The infusion of crushed esparto sprigs in boiling water is very effective in treating kidney stones.
Poaceae	Triticum durum L.	Seeds.	Bechic, laxative, astringent, caring for skin and boils, pectoral.	Wheat is used in several forms, pancake, semolina, or soup in the treatment of gastric pain, anemia and fractures.
Punicaceae	Punica granatum L.	Bark, fruit, flowers.	Vermifuge, healing, haemostatic, antidiarrheal, relieve painful menstruation.	A decoction of the barks where the fruit powder is used against diarrhea and gastrointestinal diseases.
Ranunculaceae	Nigella sativa L.	Seeds	Analgesic, antiseptic, antispasmodic, appetizer, carminative, digestive, diuretic, expectorant, febrifuge, galactagogue, vermifuge.	The use of nigella is very effective against the flu by inhaling the crushed seeds.
Rhamnaceae	Rhamnus alaternus L.	Leaves, stems, twig bark.	Astringent, laxative, purgative, effective against jaundice, hepatic.	Alterne is used as an infusion to treat jaundice (jaundice) and abdominal pain.
Rhamnaceae	Ziziphus lotus L. (Desf.)	Leaves, fruit.	Anti-inflammatory, diuretic, sedative, emollient, tonic.	Dried leaf powder, moistened with water, is applied as a poultice against boils and abscesses. Jujubes, associated with rush fruits, corn style, couch grass and prickly pear flowers, are used against kidney stones.
Rosaceae	Prunus dulcis (Mill.) DA Webb	Fruits.	Aperitif, anti-dandruff	Fruit powder mixed with milk as a poultice to improve facial skin.
Rosaceae	Prunus armeniaca L.	Fruits, barks, seeds.	Antiasthenic, antianaemic, appetizer, tonic, nutritive, astringent, laxative.	The ripe fruit is eaten to treat constipation, anemia and weakness.
Rosaceae	Prunus domestica L.	Leaves, fruit.	Softener, anti-anaemic, depurative, digestive, laxative, emollient, diuretic, astringent.	Prune (dried fruit) compote is an excellent remedy for constipation, especially if taken in the morning on an empty stomach.
Rosaceae	Pyrus communis L.	Fruits.	Anti-anaemic, astringent, depurative, tonic, anti-inflammatory, nutritious.	The fruits are eaten to treat kidney ailments.
Rutaceae	Ruta chalepensis L.	Leaves, roots.	Antispasmodic, anti-inflammatory, diuretic, emmenagogue, sedative, sudorific, vermifuge.	The root, in decoction, is used against stomach-ache, affections of the respiratory system and diseases of the liver.
Salicaceae	Populus alba L.	Buds.	Disinfectant, treatment of kidney and bladder ailments.	Poplar wood is used for heating (rural area).

Solanaceae	Capsicum annuum L.	Fruits.	Antiasthenic, aperitif, carminative, tonic, rubefacient.	Infusion of the seeds in boiling water for 24 hours is used to strengthen hair and eliminate dandruff.
Thymelaceae	Thymelaca hirsuta Endl.	Leaves.	Purgative, vermifuge.	The leaves, in decoction, are used against urinary ailments and kidney stones. The use of this plant is limited in external use, it consists in mixing the crushed leaves with olive oil to treat burns and scabies.
Vitaceae	Vitis vinifera L.	Leaves, fruit.	The leaves considered the remedy for menopause and uterine bleeding.	A gargle by infusion of the leaves against inflammation of the throat and stomatitis.
Zygophyliaceae	Peganum harmala L.	Seeds, leaves.	Rue is used as an antirheumatic.	The seeds are used in powder form in the treatment of rheumatic pain, back pain and haemorrhoids.
Zygophyliaceae	Zygophyllum album L.	Aerial part.	The plant is used as an analgesic and wound healing.	The white Zygophylle is a disinfectant used for the body care of infants.

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PHYTOCHEMICAL STUDY OF MEDICINAL PLANT « THAPSIA GARGANICA »

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Abstract: The plant Thapsia garganica belongs to the family Apiaceae and is widespread in the Mediterranean basin. It is considered one of the most famous medicinal plants since ancient times. We started this work with the methanolic extraction of all the components contained in the plant in order to obtain a crude extract, which we separated with different polar solvents "petroleum ether, chloroform, and ethyl acetate and n-butanol"

The flavonoids were evaluated by the method of aluminum chlorides $AlCl_3$, the content is estimated at 5.6009 mg EQ/g dry matter in the crude extract.

We extracted the essential oils using a Clevenger device, and then we studied the antimicrobial and antioxidant activity of all extracts and compared them among themselves.

Key words: medicinal plants, antioxidant activity, antibacterial activity.

1. Introduction

Medicinal plants have discovered and used in traditional medicine since prehistoric. they are a rich source of bioactive phytochemicals or vital nutrients. The widespread of plant medicine is attributed to several factors such as various claims on the efficacy or effectiveness of plant medicines, high cost and side effects of most modern drugs, and a movement toward self-medication [1].

The Apiaceae is a plant family comprising at the present time 466 genera and about 3800 species [2]. Commonly known as the celery, carrot or parsley family consists of dicotyledonous plants characterized in particular by their typical inflorescence [3].

Thapsia is a genus belonging to the Apiaceae family they are flowering plants with 41 species native to Africa, Asia and Europe. Thapsia garganica is a widespread species in Algeria, best known for the use of its roots in cooking and traditional medicine [4]. It is a wide

spread species in the Mediterranean basin, which inhabits roadsides and fields [4].

This perennial plant has a striated, glabrous stem, branched in its upper part, reaching 0.90 to 1.40 m in height. The leaves are green, hairless. The primordial leaves are small, elliptical and entire, the following ones are palmatilobed. The leaves at the base of the stem are large, 2-3 pinnately shaped, the upper ones are reduced to a wide sheath. The root is large, blackish on the outside, white on the inside.

The inflorescence is a large, compound umbel with 15-20 rays, bearing yellow flowers. The involucre and involucelle are absent. Umbellules are globular in shape. The fruit is elliptical, dorsally compressed, 10-15 by 20-25 mm, with notches more or less wide at the top and at the base.

Lateral wings very developed, shiny, straw yellow, finely streaked [5,6]. *Thapsia garganica* is a Mediterranean plant. It is present in Morocco, Algeria, Tunisia and Libya [7], but also in Turkey, Spain, Portugal, Italy and Greece.

Thapsia garganica it is a perennial plant, with an erect, branching flowering stem, reaching a height of about 1.50 meters. It has large leaves in tufts, very indented, linearly divided with the stem. The flowers are small, yellowish, arranged in large, nearly spherical umblas.

The fruits are oval, reach more than 2 cm in length, wide-winged. The roots are rhizome-shaped Flowering occurs between April and July. Extracts from the roots of *Thapsia garganica* used for the treatment of lung diseases, colds and relief of rheumatic pain [4].

They contain strong irritants to the skin, constituting an important cause of contact

dermatitis, which manifests itself by erythema, itching and the formation of small vesicles.

The main active compounds in the majority of root extracts responsible for these effects identified by Christensen & al. (1982) [8] as sesquiterpenes known as thapsigargine and thapsigargicine.

A study on the cytotoxicity of tapsygargin and phenylpropanoid esters isolated from *Thapsia garganica* flowers conducted by [9].

The tests done on leukemia, and breast cancer cell. IC50s revealed that thapsigargine has higher cytotoxic potentials compared to phenylpropanoid esters.





Fig. 1. Illustration of Thapsia garganica (left, plant habit and right, inflorescences), photos K. Rebbas.

2. Vegetable material

The aerial parts of the studied plant collected in June (Bejaia, Algeria) and identified by Pr K. Rebbas, a botanist from the University of M'sila, The choice of this plant is based on its use in traditional Algerian medicine. After harvesting, the collected plant material dried at room temperature and protected from light.

The resulting dry matter reduced to powder using an electric grinder. The latter kept in closed glass jars and stored away from light.

3. Extraction of essential oil

The aerial parts of the plant are subjected to hydrodistillation (for 3 hours) using a "clevenger" type extraction device, the operation

consists of immersing a quantity of the plant mass (100g) in a glass flask. (6 liter) containing a sufficient quantity of distilled water without completely filling the flask.

The mixture is brought to a boil using a heating mantle. The vapors charged with the essential oil pass through the refrigerant where the condensation will take place. Due to the difference in density, the oil floats on the surface of water and it is recovered, and then dried by a desiccant (sodium sulfate) to remove the little water likely to have been retained in the oil.

The essential oils obtained are stored in bottles protected from light and at a temperature of $4^{\circ}C$.

4. Preparation of the extracts

Thapsia garganica powder was soaked in 70% aqueous-methanol. The extract was filtered. This procedure was repeated twice on the residue using 80%, 90% aqueous –methanol respectively to obtain the last filtrate. The first and the last filtrates were combined then the methanol was removed under reduced pressure on a rotavapor below 45°C.

Crude extract (CrE) was subjected to fractionation using liquid-liquid extraction. CrE was successively extracted with different solvents of increasing polarity: petroleum ether for defatting, dichloromethane for aglycone flavonoids extraction and n-butanol for glycoside flavonoids extraction.

The obtained organic layer of each partition was evaporated under reduced pressure on a rotavapor below 45°C to dryness and to afford petroleum ether, dichloromethane and n-butanolic fractions coded as PE, DE and BE, respectively.

5. Determination of the total flavonoid content

The quantification of flavonoids carried out by a method based on the formation of complexes between phenolic compounds and aluminum trichloride [10]. The content of total flavonoids in the extracts of the plant studied was determined according to the method described by Ayad & al. (2017) [11]. One milliliter of each extract dissolved in methanol (1mg / ml) added to 1ml of AlCl₃ solution (2% in methanol).

The mixture stirred vigorously and the absorbance read after 10 minutes at 430 nm. A calibration curve produced by quercetin at different concentrations, performed under the same operating conditions as the samples.

6. Antioxidant activity in vitro

Antioxidants sometimes called "free radical scavengers" are all molecules capable of inhibiting production in low doses, limiting the proliferation or destruction of reactive oxygen species. They prevent cell damage from highly reactive and unstable molecules called "free radicals" [12, 13].

The balance between antioxidants and free radicals in our bodies is important for health. If not controlled, free radicals lead to cell damage associated with a variety of chronic diseases. The sources of antioxidants can be natural or

artificial. The body also produces some antioxidants known as endogenous antioxidants in contrast to antioxidants that come from outside the body called exogenous [12].

Indeed, DPPH characterized by its ability to produce stable free radicals; this stability is due to the delocalization of free electrons within the molecule. The presence of these DPPH • radicals gives rise to a dark violet color in the solution. has been proven that the reduction of DPPH • radicals by an agent antioxidant causes discoloration of the solution, and the color change can be followed spectrophotometrically at 517nm and in this way the antioxidant potential of a substance or a plant extract can be determined [14].

DPPH 2,2 – Diphenyl – 1 – picrylhydrazyl ($C_{18}H_{12}N_50_6$; Mr: 394.33), is dissolved in absolute methanol (4 mg / 100ml).For the test, the samples were prepared by dissolving in absolute methanol. For all extracts, solutions in absolute methanol were prepared. These solutions, called stock solutions, will then undergo dilutions in order to have different concentration.

The protocol used for the evaluation of the scavenger effect of plant extracts against the DPPH radical is that of "Cuendet" with a small modification [15].

50 μ l of the test solution are introduced into dry and sterile tubes, 1250 μ l of the DPPH solution are added. After vortexing, the tubes placed in the dark at room temperature for 30 minutes. For each concentration, the test repeated 3 times. The reading taken by measuring the absorbance at 517 nm by a spectrophotometer. The negative control is composed of 1250 μ l of the methanoic solution with DPPH and 50 μ l of methanol.

7. Expression of results

To obtain the effective concentration, which reduces the initial concentration of DPPH by 50%, the results expressed as antioxidant activity. The antioxidant activity, which expresses the capacity to scavenge the free radical, estimated by the percentage of discoloration of the DPPH in solution in methanol. The "AA%" antioxidant activity given by the following formula:

$$\begin{array}{l} AA~\% = 100 - \{[(Abs_{test} - Abs_{blanc}) \times 100]/\\ Abs_{control}\} \\ Inhibition~\% = (Abs_{control} - Abs_{test})/Abs_{control} \times \\ 100 \end{array}$$

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AA: Antioxidant activity.

Abs: Absorbance at the wavelength of 517 nm.

The results expressed as the mean of two measurements ± standard deviation. The IC 50 value was determined for each extract, is define as the concentration of the substrate, which causes the loss of 50% of the activity of DPPH (color).

8. Antibacterial activity

The aim of this work is to evaluate the antibacterial activity of the essential oil and the other four extracts (n-butanol extract, ethyl acetate extract, chloroform extract and petroleum ether extract) of *Thapsia garganica* against bacteria.

a. Diffusion method in solid medium

The study carried out by the diffusion method, which is initially designed for antibiotics (antibiogram), but substituting the antibiotic discs with others impregnated with (n-butanol extract, extract of acetate ethyl, chloroform extract, petroleum ether extract) of *Thapsia garganica*. This method consists of depositing sterile

Wattman paper discs N° 3 and 6 mm in diameter impregnated with 10 μl of each extract with

different concentrations, the discs placed at the surface of the petri dish in the presence of discs soaked with an aqueous solution (negative control) placed at the agars seeded with the germ to be tested and to measure the diameters of inhibition in millimeters (mm) after incubation.

b. Preparation of solutions

We dissolved each of the essential oil extract and four other organic extracts so that:

- The different organic extracts of the studied plants were dissolved in DMSO and dilutions were made to obtain concentrations of 0.2 g/ml for each tested extract.
- The essential oil extract obtained from distillation was dissolved in DMSO in order to obtain a concentration of 0.1mg/ml.

c. Microbial strains used

The strains used to detect the antibacterial activity of *T. garganica* extracts belong to four genera of microorganisms, which are reference strains of the American Type Culture Collection (ATCC), these are: *Staphylococcus aureus* (ATCC 25293), *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), *Enterococcus*(ATCC 51299).

Table 1: Microbial strains used

Gram+	Gram-
Staphylococcus aureus (ATCC 25293)Enterococcus (ATCC 51299)	 Escherichia coli (ATCC 25922). Pseudomonas aeruginosa (ATCC 27853).

d. Preparation of the inoculum

First, a bacterial suspension is prepared from a pure and young culture (18 hours old). This inoculum used to inoculate Mueller Hinton agars poured into petri dishes to a thickness of 4 mm and then dried in an oven at 37°C before use. Inoculation carried out by swabbing, from the freshly prepared inoculum.

It consists of dipping a sterile cotton swab in the suspension then rubbing it, after having wrung it inside the tube, three times over the entire agar surface so the inoculation carried out by swabbing, from the inoculum to form tight streaks, rotating the dish approximately 60° after each application to obtain an even distribution of the inoculum.

Pre-sterilized watman paper discs 6 mm in diameter placed on the surface of the inoculated agar after having been loaded with $10~\mu l$ of

extract. After 24 hours of incubation at 37 $^{\circ}$ C, the diameter of inhibition is measured.

9. Results and discussions

9.1. Total flavonoid content results

The determination of the flavonoids was carried out according to the trichloride method of aluminum $AlCl_3$ and the standard was quercetin, The flavonoid content was calculated for the crude extract which is expressed in μg EQ / mg of extract.

Before proceeding to the determination of the content of flavonoids we have established a calibration curve using quercetin as a reference compound. The flavonoids were evaluated by the method of aluminum chlorides $AlCl_3$, the content is estimated at 5.6009 mg EQ / g dry matter in the crude extract.

9.2. Result of antioxidant activity evaluation in vitro

Trapping of the 2-diphenyl-picrylhydrazyl radical (DPPH):

The DPPH test has attracted a lot of attention due to its speed, sensitivity and reproducibility. The results obtained in this study reveal a proportional relationship between the concentration of petroleum ether / acetate ethyl extracts as well as the crude extract and the BHT standard with the percentage inhibition of the DPPH radical.

The IC 50 of the different extracts are compared with the BHT standard. The IC50s in the order of antioxidant power were: petroleum ether (42,2938144 mg / ml) > acetate ethyl (0,67893784mg / ml> BHT (0.0059 mg / ml). Depending on the results we have obtained, we can predict that flavonoids are first class antioxidant agents.

9.3. Antibacterial activity

The results regarding the Antibacterial activity of the essential oil and the four extracts petroleum ether, chloroform, acetateethyl, n-butanol of "*Thapsia garganica*" are indicated in Table 2. The results obtained from disc diffusion method indicated that the essential oil and the four extracts didn't show any Antibacterial activity against all microorganisms tested.

The diameters of inhibition of *Pseudomonas* aeruginosa (ATCC 27853), *Staphylococcus* aureus (ATCC 25293), *Escherichia coli* (ATCC 25922), *Enterococcus*(ATCC 51299), was 6 mm and this indicates the insensitivity of the bacteria against both the essential oil and the extracts.

Table 2. Antibacterial activity of essential oil and the extracts of Thapsia garganica

Diameters of inhibition zones (mm)									
Bacterial strain	N-butanolextract (NB)	Acetate ethyl extract (AC)	Chloroform extract (CH)	Petroleum ether extract (PE)	Essential oil extract				
Escherichia coli (ATCC 25922	6	6	6	6	6				
Staphylococcus aureus (ATCC 25293),	6	6	6	6	6				
Pseudomonas aeruginosa (ATCC 27853)	6	6	6	6	6				
Enterococcus	6	6	6	6	6				

10. Conclusion

Thapsia garganica is one of the most important plants in traditional and modern medicine, it belongs to Apiaceae family. It was devoted to studying the antibacterial and antioxidant properties of this specie, as well as the discover of its polyphenolic content.

Quantitative determination of total flavonoids by the alimunium chloride $AlCl_3$ reagent revealed that T. garganica is rich in flavonoids.

The study of the antibacterial activity by the method of diffusion on agar medium showed that the plant did not exhibit an antibacterial activity on most of the strains used.

The study of the antioxidant activity showed that the plant rich of antioxidant elements, it_could be

used as a source of natural polyphenols and antioxidants.

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PRELIMINARY RESEARCH ON CONSUMER ACCEPTANCE OF 6% REPLACEMENT DEGREE PROTEIN FISH POWDER IN SNACKS

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Abstract: The paper presents the results of preliminary investigations on the acceptability of snacks with the addition of 6% protein flour from salmon. Protein flour was obtained by hydrolysis of waste from a salmon processing factory in Norway. The sncaks type products were made in 2 variants, with and without cumin (3%). The 12 respondents were selected on the basis of a questionnaire, based on principles such as: non-smokers, without medical conditions that may influence the answer. The results showed an overall degree of acceptability of 6.92 from a maximum of 9. The researches are part of the project activities of contract no 245/2021, project acronym SUMAFOOD (SUSTAINABLE PRESERVATION OF MARINE BIOMASSES FOR AN ENHANCED FOOD VALUE CHAIN), stage 2, 2022.

Keywords: fish protein powder, snacks, consumer acceptance, total acceptability

1. Introduction

Globally, fish processing industries generate and dispose a large volume of waste (skin, wings, head, viscera, scales and roe) each year, estimated up to 60% of the harvested biomass [1]. According to Kang et al. 2018 [4], Nazeer et al. 2011 [7] and Šližytė et al. 2009 [9], the waste obtained from fish factories can be converted to value-added products such as fish protein hydrolysate (FPH) with the addition of various proteolytic enzymes. FPH from fish processing waste, including skin, heads, muscles, bones, etc., are a rich source of protein, amino acids, peptides and antioxidants and has been found to possess functional and bioactive peptides [2, 3, 5, 6, 10].

A moderate inclusion of FPH in aqua-feeds has the potential to improve growth, feed utilization, immune functions and disease resistance of fish. Production of FPH, targeted to more precise molecular weight ranges, has superior functionalities that are in high demand.

Salmon protein hydrolysate (SPH) is made from fresh by-products from farmed salmon that are minced and acidified to hydrolyse proteins into peptides and free amino acids [8]. In the frame of the ERANET BLUE-BIO project, contract no. 245/01.10.2021, the possibilities of using fish protein powder (obtained by hydrolysis of salmon waste from a processing unit in Norway) were studied, both from technological and from consumer acceptability point of view. The research is part of the broader framework of systemic approaches to a circular economy in the field of aquatic bioresources, the capitalization of algae in the bakery industry being another important component in the "BlueBIO" paradigm addressed in the ERANET 2021 call.

2. Materials and methods

To test the reaction of consumers regarding the new ingredient, snacks with a diameter of about 60 mm and a thickness of approx. 2 mm were obtained. The degree of replacement of wheat flour in the recipe was 6%, making 2 variants:

- with the addition of 3% cumin seeds (to reduce the after taste);
 - without the addition of cumin seeds.

The answers of a number of 12 selected respondents, non-smokers and without medical conditions, aged between 22 and 60 years, of which 45% were men and 55% women, were registered.

The registration of the answers was done using the google docs platform.

3. Results and discussion

Figure 1 shows an average value in terms of the colour of the outer surface. 7 of the 12 persons appreciated the colour intensity of the product as being grade 3 out of 5. 3 themselves appreciated the colour with grade 2, and 2 respondents with grade 4 out of 5.

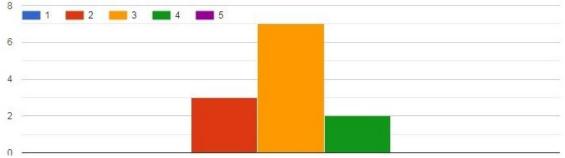


Fig. 1. Crust colour intensity of the salty snacks with 6% hydrolysed salmon fish protein

Regarding the intensity of the bitter aroma (figure 2), 7 respondents appreciated with grade 1, the grade indicating an imperceptible aroma, 3

appreciated with grade 2 and 2 respondents with grade 3 out of 5.

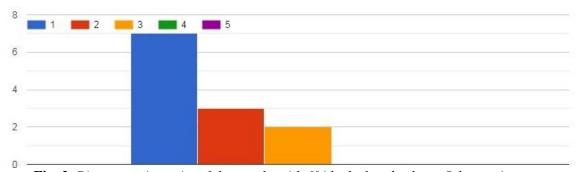


Fig. 2. Bitter taste intensity of the snacks with 6% hydrolysed salmon fish protein

The salty taste (figure 3) was perceived as imperceptible by 7 respondents, being marked with a grade of 1 out of 5, 1 respondent

appreciated the flavour with a grade of 2 and 3 with a grade of 3 out of a maximum of 5.

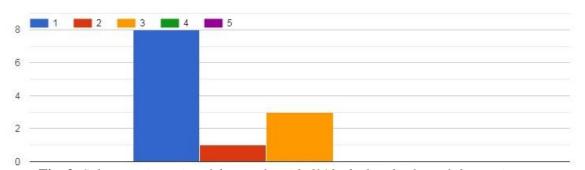
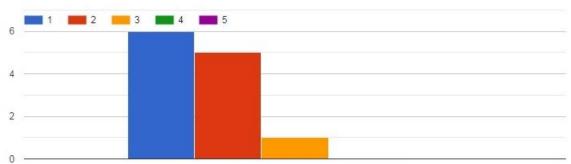


Fig. 3. Salty taste intensity of the snacks with 6% hydrolysed salmon fish protein

The sour taste (figure 4) was appreciated by 8 respondents as being imperceptible, 1 respondent appreciated with grade 2, and 3

persons appreciated with grade 3 out of 5 the intensity of the sour aroma.



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Fig. 4. Sour taste intensity of the snacks with 6% hydrolysed salmon fish protein

The specific aroma (figure 5) was appreciated with grade 1, by 3 respondents, by 2 with grade

2, by 4 respondents with grade 3 and 3 appreciated with grade 4 out of 5.

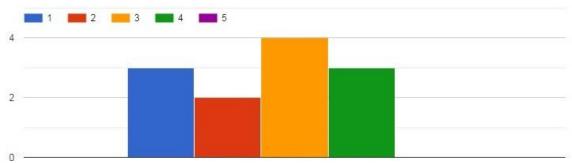


Fig. 5. Specific aroma intensity of the snacks with 6% hydrolysed salmon fish protein

Regarding the after taste, 4 respondents rated with a grade of 2, 6 persons with a grade of

4 and 2 respondents rated with the after taste with a maximum grade of 5.

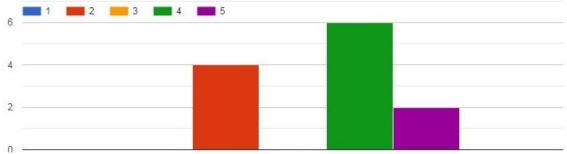


Fig. 6. After taste intensity of the snacks with 6% hydrolysed salmon fish protein

Also, the total acceptability of the products was studied by a hedonic test in 9 points (figure 7 and 8), where out of the 12 respondents, 1 appreciated with a grade of 4 (I slightly dislike it), 3 appreciated with a grade of 6 (I slightly like

it), 4 respondents rated it with a grade of 7 (I like it moderately), 3 people rated with grade 8 (I like it very much) and one person appreciated with the highest grade, 9 (I like it extremely).

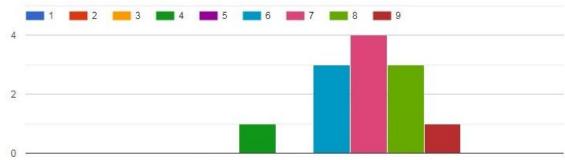


Fig. 7. Total acceptability of the snacks with 6% hydrolysed salmon fish protein

The total acceptability rate was measured by using the following 9 level hedonic test, where: 9 - I like it extremely; 8 - I like it very much; 7-I like it moderately; 6 - I slightly like it;

5 – indifferent; 4 – I slightly dislike it; 3 – I dislike it moderately; 2 – I dislike it very much; 1 – I dislike it extremely).

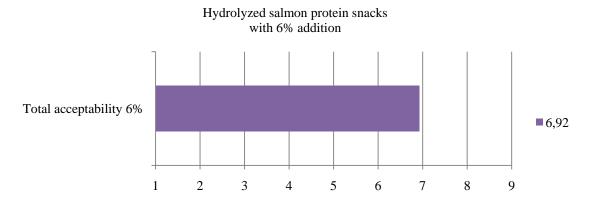


Fig. 8. Total acceptability of the snacks with 6% hydrolysed salmon fish protein



Fig. 9. Snacks with 6% hydrolysed salmon fish protein with and without 3% cumin seeds addition

Figure 9 shows the products obtained, with and without the addition of cumin seeds (3%).

Conclusions

The bakery technology is an easy to use environment for capitalizing on hydrolyzed fish protein flour, ensuring an increase in the content of proteins and minerals.

Following the research carried out on the panel team, it is found that there is an average acceptance of snack products with the addition of 6% hydrolyzed fish protein.

Future researches aim to increase the degree of replacement of wheat flour with hydrolyzed fish powder, as well as the introduction of new ingredients in the recipe, such as flaxseed, garlic powder, etc. to increase consumer acceptance regarding after taste.

Acknowledgement

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EFFECT OF THE INCORPORATION SPIRULINA ON THE PHYSICOCHEMICAL PARAMETERS OF A GRAPE JUICE

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Abstract: Our study focuses on the valuation of Spirulina, which can contain up to 70% of protein, in the manufacture of a dietary product. It aims to elaborate a hyper-protein energy drink of grape juice having a significant nutritional intake .Our study aims to show the effect of the incorporation of spirulina on the physicochemical parameters of a grape juice at different doses (0.1g/l - 0.3g/l - 0.5g/l - 0.7g/l) compared to a control drink and evaluation of its nutritional value. Several analyzes were carried out The Physicochemical analyzes - acidity, Brix, pH, and density with reference to Algerian standards, as well as sensory analyzes; stability testing. The statistical analysis (Newman-keuls test) showed that they are not significantly different between all parameters at the probability 0.05 probability level. The physico-chemical analyses obtained from different products are in agreement with Algerian standards. Indeed, the juice has a certain richness in proteins (31%); it is therefore considered a high protein product. A good microbiological quality of the juice was revealed. The F3 formulation (0.5g) is judged to be good, and well appreciated by the tasting panel. Its stability test for 4 weeks at 22°C showed that formulation F3 (0.5g) is a stable juice. There is a significant effect (p < 100) of the dose of spirulina incorporated on the parameters studied (Acidity, pH, Density, refractive index). The enrichment of grape juice by spirulina allowed us to obtain a dietetic grape juice, a stable and hyper protein with a protein level of 31%.

Keywords: Enrichment, Grape juice, Spirulina, Stability test, Nutritional value.

1. Introduction

The food industry has shown interest in the application of ultrasound to improve the chemical, physical properties of various foods (Higuera-Barraza et al., 2016). Ultrasound-assisted extraction (UAE) is efficient in the disruption and extraction of algal cells without the addition of beads (for milling) or chemicals, with less energy consumption at relatively lower

temperatures leading to less thermal protein denaturation. Also, UAE offers a great reduction in time and solvent consumption (Picó, 2013).

Many authors studied the effect of ultrasound on protein extraction. The use of ultrasound increases the concentration of proteins of Spirulina. Another study shows the positive effect of sonication time and sonication energy on recovery protein yield. The application of Surface Response methodology in the optimization of analytical procedures is very beneficial due to the generation of a large amount of information from a small number of experiments and the possibility to evaluate the interaction effect between variables on the response (Bezerra et al., 2008).

Spirulina is a great source of natural protein (about 60% digestible proteins). It is low fat, low calorie, cholesterol-free source of protein with all amino acids, phytonutrients, antioxidants, carbohydrates, mucopolysaccharides, itamins, and trace minerals. It is a ubiquitous organism that was used as food in Mexico 400 years ago during the Aztec civilization (Al-Dhabi, 2013).

Spirulina is a freshwater cyanobacterium characterized by a blue-green color due to the presence of a rare protein pigment. It is considered an unconventional food resource that can contain up to 70% of its dry weight, twice as much soy protein, and three times more than beef (Higuera-Barraza et al., 2016).

The spirulina protein supplies the body with almost all essential amino acids, making it an essential food for vegetarians.

Spirulina is used as a dietary supplement, which can be used with all the food and drink we consume. We tried to develop spirulina in the manufacture of a food product namely high protein grape juice.

Fruit juices, derived from a wide range of fruit, including oranges and another citrus, apple, grape, pineapple, and mango, often result from the fruit juice blend of several species. Usually, fruits are processed into concentrates near their harvesting place and then shipped to a conditioner.

The grape juice is an unfermented but fermentable juice, intended for consumption directly and obtained by a mechanical process from sound and ripe grapes exclusively preserved by a physical process.

The juice may have been concentrated and reconstituted with suitable water to preserve the essential compositional factors and the quality of the juice (Picó, 2013).

Our research consists of a comparative study between two grape drinks (one enriched with spirulina and another non-enriched drink) and an evaluation of its nutritional value with enrichment with different doses of spirulina (0.1 g /L - 0.3 g /L -0.5 g/L - 0.7 g/L). The physicochemical analysis is conducted while respecting its organoleptic properties to get at the end an energy drink with a high nutritional intake.

2. Materials and Methods2.1. Material study

The process water was removed aseptically using 3 sterile 225 ml glass bottles of the storage tank. The valve was well cleaned with a disinfectant (alcohol 90°) and then rinsed. After letting the water run for a few minutes, the bottles were filled and closed.

- 30 g of grape concentrate was taken aseptically under hygienic conditions and introduced into 3 sterile vials.
- 30 g of sugar were collected aseptically in ultrahygienic conditions and introduced into 3 sterile vials.
- Spirulina powder (Vita Spirulina) N ° lot:. 011,413,790 (Spirulina available is in the form of original high-quality Morocco powder), this powder is kept away from moisture. It is rich in bioactive components and nutritional substances. This gives it a high potential for use, especially in the diet, as a supplement rich in protein. The nutritional properties of Spirulina make it a food source that deserves special attention for its development.
- Semi-finished product (before pasteurization grape juice).
- Finished Product: these are the bottles cardboard Tetra Pak capacity of 11L which are the subject of our study.

2.2. Sampling

The study focused on five different productions, five products were the subject of our study namely grape concentrate, water, the semifinished product (before pasteurization), the finished product, and the finished product enriched with spirulina. We have prepared a juice "grape nectar" enriched using different doses of spirulina. For this drink, we prepared four samples with different doses of spirulina. The composition of each sample is as follows:

- -specimen 1: contains spirulina 0.1g in 1 liter of grape nectar.
- -Specimen 2: contains spirulina 0.3 g in 1 liter of grape nectar.
- -Specimen 3: contains spirulina 0.5g in 1 liter of grape nectar.
- -Specimen 4: contains spirulina 0.7g in 1 liter of grape nectar.
- -Specimen Preparation

To prepare these 4 samples, are mixed:

- 1 liter of juice "grape nectar"
- Spirulina at the different doses

- Mix the 500ml grape juice with the selected spirulina dose in a beaker and shake well for 5mins with an agitator.
- Reverse this mixture into the remaining 500ml of grape juice, and mix it again for a few minutes with a stirrer.

2.3. Methods of physicochemical analysis2.3.1. Physicochemical Analyses performed on water

The physico-chemical analyzes are carried out on two types of water: water boiler, and that of the process.

- pH Principle: The pH was measured directly using a pH meter. Result: Direct reading of pH on the pH meter.
- -Title gauging water (TH): according to the standard(Bezerra et al., 2008).

Principle: Total hardness or title of a hydrometric water corresponds to the sum of concentrations of metal cations. In most cases it is mostly due to ions Ca^{++} and Mg^{++} (METHOD complexometrically). If the resulting solution is blue, so TH=0. If the resulting solution is violet, proceed to the titration Solution Ethyl Diamine TetraAcetic acid (E.D.T.A) 0.02 N to turn blue.

TH = 1000. C. V1/V2

The total concentration of Ca^{++} and Mg^{++} expressed in mmol/ 1

C : Concentration mol / 1 of the solution of 0.02N ETDE

V1: Volume in ml of the solution E.D.T.A,

V2 : Volume in ml of the sample (100 ml),

Conversion : $0.1 \text{ mmol} / 1 = 1 \degree \text{F}$

TH ($^{\circ}$ F) = V1

-Full Title and alkalinity alkalinity water TA and TAC [4][8] (AFNOR., 1986), principle An alkaline water is estimated by the acidimetry carbonates of HCO₃

- Present therein Expression of results

TA = V1.5°F then: TA = V1.10°F

TA is expressed in meq is converted to level French: $1 \text{meq} = 5^{\circ}\text{F}$

V 1: volume of H₂SO₄ used for titration.

TAC= 2V. 5° F so: TAC=V.10°F

V: volume H_2SO_4 versed in $V_1 + V_2$ solution.

Chlorides [3](NA 6917); principle Chlorides are determined by a solution of chromate in the presence of silver nitrate potassium. The reaction is indicated by the appearance of red color characteristic of AgCl.

$$(CL^{-}) V_{\bullet} = 100 \text{ mg/l}$$

V : volume AgNO₃ paid.

2.3.2. Physical and chemical analyzes of the concentrate

The physicochemical analysis is first performed at dilutions to measure the degree of Brix and Titratable acidity.

Brix (refractive index) (SI 2173 / EN 12143).

Principle, The degree of Brix is measured by refractometer. The reference temperature is 20°C. Brix value in a program is given by the following formula:

Brix = lectue.4 (g/kg)

Titratable acidity according to the standard (NA 691 / EN 12147), principle, Acidity analysis by titremetric method using a known normality basis.

2.3.3. Physico-chemical analysis performed on the semi finished and finished products.

There are four parameters to analyzes: pH, acidity, Brix, and the density. There are performed only for the finished product. Density (Al-Dhabi, 2013).

Principle, Determination of the density and the corresponding temperature of the product to be inspected by direct reading.

2.3.4. Physico-chemical analysis performed on Spirulina

Water content: The standard NA / 1133/1990) (Benahmed Djilali,2012).

a) Principle: The method consists of a practical reference steaming at atmospheric pressure, at a temperature 130-133°C under defined operating conditions, the mass loss observed is equivalent to the amount of water present in the product.

The potential of hydrogen (pH): According to NF V05-108 standard in July 1970. The total protein content: According to the standard (Bezerra, 2008).

The total lipid content: According to the (Codex Alimentarius ,1992), AFNOR NFV03-713 (1984).

The total carbohydrate content "G" in g per 100 g of dry product is calculated by difference:

$$G = 100 - (H\% + CB + C\%\%\% + L + P\%)$$

Along With:

H: moisture content (% of dry product),

CB: ash content (% of dry product),

P: total protein content (% of dry product),

L: total lipid content (% of dry product).

Energy value: The determination of protein, fat and carbohydrates content allowed us to calculate the energy value for each drink prepared according to the formula:

Energy value in Kcal = 4 carbohydrate + 4 proteins + 9 lipids

Determination of total ash: According to Standard No. 22,97.07

2.3.5. Sensory analysis

The choice of the best formulation is based not only on the nutritional qualities but also on the organoleptic qualities which led us to perform the sensory analysis.

Sensory analysis therefore aims to describe the sensory characteristics of the products in an objective and quantifiable way according to defined criteria appearance, texture, flavor and aroma (ISO 6222, 2011)

A questionnaire was distributed to people from different backgrounds (academics, officials and employer of the manufacturing unit Vita juice).

For each sample, the tasters noted the level of overall acceptability using a structured 4-point scale. As the sensory evaluation is focused on the general acceptability, Note 1 corresponds to poor quality product on the taste level, odor and appearance and Note 4 corresponds to a good quality product.

2.3.6. Juice stability test

The stability test was conducted on the best chosen formulation in sensory analysis (F3 juice). This juice was prepared and analyzed the same day and then analyzed after incubation at 22 ° C for 4 weeks.

The main interest is to observe the evolution over time to estimate a deadline for optimal use and to study the stability of this juice and its physicochemical, organoleptic and microbiological quality.

2.3.7. Dissolution of phycocyanin

Based on its pharmacological properties, phycocyanin, is studied as an active substance, deserves attention from the point of view of release phenomena from tablets immersed in various liquid media (distilled water, HCl 0.1 N and phosphate buffer solution pH 6.8).

The dissolution test consists of placing each tablet in 500 ml of dissolution liquid in the dissolution apparatus (Figure 1) equipped with paddles operating at 50 r.p.m and the temperature of the medium is set at 37 ± 0.5 °C (Sriamornsak, 2007) .

Phycocyanin content is calculated at different time intervals using the colorimetric method described by Jourdan (Jourdan, 2012) which involves measuring absorbance at 615 and 652 nm.

Phycocyanin concentration was obtained by tuning equation 15:

$$M_{ph_t} = 1,873 * (DO_{615} - 0,474 * DO_{652}) * \frac{DIL}{C}$$

Equation 1

DIL: Dilution factor;

 M_{ph} : Initial phycocyanin concentration (%);

 DO_{615} , DO_{652} : The optical density (OD) measured at wavelength 615, and 652 nm respectively (m-1);

The concentration of dry spirulina soaked in water around 4%.

The results obtained, percentage of dissolved phycocyanine compared to the initial quantity in the tablets are represented as a function of time in the form of a graph.

2.4. Statistical analyzes of the results

For the interpretation of the results obtained, the data were subjected to a statistical analysis:

An analysis of variance to determine the effect of the doses on the studied parameters and their interactions. The significance of the results is expressed according to their probability.

the classification of homogeneous groups was carried out by the test NEWMAN and KEULS which makes it possible to classify the values at a threshold of 5%.

A correlation analysis was performed between the physicochemical parameters of the samples. These statistical analyzes were performed using STATISTICA software. Version 6.

3. RESULTS

3.1. Results of physico-chemical analysis

3.1.1. Process Water

Physicochemical analyzes were performed to verify the effectiveness of these treatments.

Knowing that this water has a direct influence on the organoleptic quality of the product.

The results of the physicochemical analyzes of the process water are shown in table 1

Table 1. Results of	of physicoch	hemical anal [,]	ysis of the	semi-finished	product

Analyses Specimen	TH	pН	Cl-
E1 11/05/14	5,7	7,30	50
E2 13/05/14	4,9	7,25	60
E3 15/05/14	8,5	8,46	35
E4 18/05/14	19	7,55	38
E5 21/05/14	10	7,22	40
Standards	=10	7 à 8,5	Max 40 mg/1

The results of the physicochemical analysis of the process water, showed that:

The pH of the samples varies between 5 (7.22 to 8.46), which complies with the internal standards of the Vitajus unit.

TH of the four samples (E1, E2, E3, E5) is consistent with the standard except the E4 is superior to the standard. This non-compliance of the TH is probably due to high concentrations of metal cations that are typically calcium and magnesium and resulting in the saturation of an ion exchange resin (softener). This latter also requires regeneration.

Chloride of the three samples (E1, E2) is greater than the standard. The presence of

excessive chloride in water promotes corrosion (when water is not charged in limestone, it is often corrosive to metal water pipes. In each case, it may follow significant cost of repairing pipes (leaks or blockages), so it is necessary to reduce the chloride concentration because it will influence the organoleptic quality of our finished product.

3.1.2. Semi finished product

The results of physicochemical analysis carried out on the semi-finished product are shown in Table2.

Table 2. Results of physicochemical analysis of the semi-finished product.

Analysis Spicemen	acidity	refractive index	pН	Density
		4.5.0	2.02	
E1	3,50	15,2	2,93	1,064
E2	3,92	15,3	3,14	1,065
E3	3,78	14,9	2,78	1,062
E4	3,50	15	2,90	1,063
E5	3,38	15,4	3,02	1,059
Standards	3,22 à 4,20 g/kg	14,6 à 15,5 Bx	2,50 à 3,20	1,058 à 1,064

The results show that the five samples conform to the standard established by the Vitajuice unit. This reflects the respect of the recipe when preparing the doses of five samples, thus the product can be pasteurized and packaged.

3.2.2. Finished product

The results of physicochemical analyzes carried out on the finished product are summarized in Table 3.

Table 3. Results of physico-chemical analysis of the finished product.

Analyzes Samples	Acidity	Refractive index	рН	Density
Control	3,64 b†	15,1 a	2,74 с	1,064
0.1	3,22 a	15,3 a	2.72 a	1,065
0.3	3,36 a	14,9 c	3.09 d	1,062
0.5	3,64 b	15 b	2,80 cd	1,063
0.7	3,22 b	15,0 a	3,18 b	1,059

The statistical analysis (Newman-keuls test) showed that they are not significantly different between all parameters at the probability 0.05 propability level there is a significant effect ($p \!<\! 100)$ of the dose of spirulina incorporated on the parameters studied (Acidity ,pH, Density , refractive index). The Brix degree of all the samples prepared varied between (3.22 and 3.64), however we can say that the Brix values of 5 samples meet standards required by J.O.R.A.

The four samples have almost the same pH ranging between (2.72 and 3.18) on average and that does not exceed the critical limits that are

between (2.50 and 3.20). The density ranging from (1.058 to 1.059), these samples have a density according to the standard which is of (1.058 and 1.064). The results obtained for acidity meet the standards required by Vitajus company, which confirms the physico-chemical quality of the finished product.

3.2.3. Spirulina

The results of physicochemical analyzes of spirulina are shown in Table 4.

Table 4. Physico-chemical analysis of Spirulina.

Tests	H%	Dry	pН	Ash content %	Protein content
Spirulina	6.14	93.86	8.03	10.79 ± 0.1	61.25±0.17
Standards	<10	>90	7-9	<10	>50

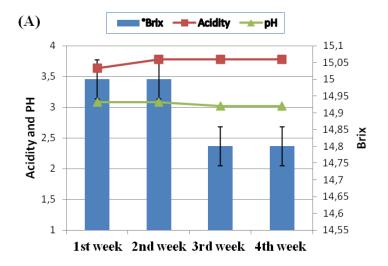
According to (AFNOR, 1986). We note compliance of all the results: pH, dry matter, moisture, ash content and protein content. For Humidity (% H) and dry matter (DM%): Moisture is a very important factor in the physical and chemical characteristics of Spirulina. There is 6.14% in the case of Spirulina. This value is very close to that found by (Norme NF EN ISO 6222, 1999) for dried products (between 4-6%). Furthermore, it should be noted that our result is lower than that obtained by (NF V03-713,1984). Which are around 13.62%. In

fact, the higher the humidity, the more microbial proliferation and less material is stored. In addition, these results show that Spirulina is very rich in dry matter with a rate of 93.86%, which corresponds to the standard announced by (AFNOR,1986) which is greater than 90%.

3.2.4. Sensory analysis

Stability test F3 juice

The results of sensory analysis are shown in Figure 1.



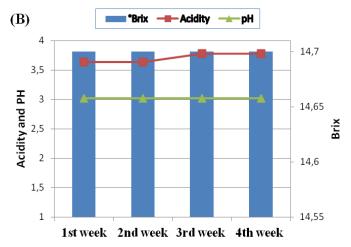


Fig. 1. Variation of stability parameters of juice enriched at 22°C for 28 days (A) and witness juice (B)

Standards : °B (14,4 - 15,5) Acidity (3,08 - 3,78) pH (2,50 -3,20)

The stability test results show that the values of the acidity, Brix, pH juice grape enriched with spirulina for 1 week 2nd week 3rd week and 4th week are in line with standard. Note that the enriched juice is stable, there is no change in the physicochemical criteria.

For the values obtained from the control juice (not enriched), they showed no changes occurred in the first and the second week, so a good juice physico-chemical quality.

By the fourth week we observe a decrease of the index of refraction to 14.8 but it remains at the norm, with a stability of pH values and acidity.

These analyses are accompanied by a taste test, and we note that there is no change in sensory quality (taste, aroma with excellent color characteristic grape juice). The results of physicochemical analysis of the 4th week of the control revealed that juice Brix values (14.5), pH (3.02) and acidity (3.78) are included in the

standards. The results of organoleptic analyses indicate that at the 4²th week the color, the taste and the smell did not undergo any modification. This affirms the Sensory quality of our juice.

3.2.5. Microbiological analyses of the formulation F3 (0.5g)

The total germs sought are germs at 30 $^{\circ}$ C, Total coliforms at 37 $^{\circ}$ C, 30 $^{\circ}$ C Yeasts and molds 30 $^{\circ}$ C.

The formulation F3 is considered stable. The absence of microbial flora in the samples treated with steam is only a confirmation of the results of the physical and chemical analyses, in the physical aspect in particular (no bending of the bottles). The pH variation does not exceed 0.5 units. This can be attributed to the quality of raw materials, the efficiency of the heat treatment applied to grape juice and juice final before steaming and the hygienic conditions in the preparation and analysis. The results shown in Table VI revealed that it is enriched juice of a

good microbiological quality. We demonstrate by the end that spirulina has not contaminated the juice.

3.2.6. Nutritional value of the enriched juice

The nutritional value is the quality of food according to the nutrients that it contains.

This test was performed on a grape juice fortified with Spirulina $(0.5g\ /\ l)$ to study its nutritional value.

The following report represents (Table 5).

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Table 5. The nutritional ve	nine oi	grupe jui	ice enricheu	will s	yu uuuu	U.J.K/	$\iota \wr \Lambda$	ccoraing	LUU	1

Determinations	Unit	Control juice	Enriched juice	Methods
Sugar content(carbohydrates)	<u></u> %	12.00	14.00	Bertrand
Lipid content	%	0.01	0.02	Petroleum ether extraction
Protein content	%	0.12	0.43	Kjeldhal
Energetic value	Kcal/100ml	48.57	57.90	Calcul

The results of the nutritional value performed on the enriched grape juice and the control, obtained from an external laboratory (LBQ).

The results of the nutritional value of the enriched grape juice showed that the protein content increased compared to the finished product (ordinary grape juice).

The results of the energy value performed on the enriched grape juice demonstrate an increase in value compared to the energy value of the unfortified juice (Finished product). From these results, we can note that spirulina influences nutritional value with the increase in protein and therefore increasing the energy value.

3.2.7. Dissolution of phycocyanin

The dissolution process is naturally influenced by the tablets' abilities to erode and disintegrate (Benahmed Djilali et al.,2011). The release rate of phycocyanin from our tablets (Figure 2) significantly distilled increases in compared to the other two liquids examined, this could be due to the molecular structure of the investigated substance and its high affinity to water (hydrophilic) (Hirata, 2000), but the low release rate (17% in 90 minutes) of phycocyanin is an important factor in terms of its therapeutic properties since a low dose of phycocyanin is more effective as an anti-inflammatory substance (Degbey, 2006).

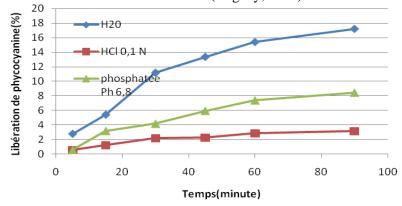


Fig. 2. Phycocyanin release rate as a function of immersion time for different media.

The results are in agreement with the investigations about different phycocyanin dissolution solutions (Jayant Mahadev, 2005). This author found that the highest concentration was obtained with distilled water and phosphate buffer solution at pH=6.8 while HCl 0.1 N leads to a negligible phycocyanin diffusion rate, around 2% (Benahmed Djilali et al.,2011). On the other hand, several studies have shown % release after 1 h of 30% for paracetamol (Parojcic,2007).and 12% for metronidazole (Limmatvapirat, 2008), These results make the tablets more interesting for melting in the mouth (neutral pH)

CONCLUSION

The results of analysis of the nutritional value of enriched juice confirmed a significant increase in protein (31%) and carbohydrates (2%), relative to the unenriched product, we also note that the energy value has considerably increased compared to unenriched finished product; it allows us to judge our grape juice as hyper protein. The stability test consists of studying the evolution of grape juice enriched over time in order to estimate a deadline for optimal use, we noticed that the grape juice is not fermented for 4

weeks at room temperature 22°C. Our results led us to choose a grape juice stable and hyper protein with a good bacteriological, physicochemical and organoleptic quality.

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ETHNOMEDICINAL USES, PHYTOCHEMISTRY AND BIOLOGICAL ACTIVITIES OF TALGHOUDA (BUNIUM FONTANESII BATT. AND RELATED SYNONYMS): A REVIEW

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Abstract: The purpose of this review is to provide comprehensive scientific information on Talghouda (Bunium fontanesii), the geographical distribution, botanical description, its chemistry, pharmacological properties of species are stated. As much data on B. fontanesii was gathered using several scientific search engines as: Google Scholar, Science Direct, PubMed, SpringerLink, Web of Science and Scopus. In this review, studies mentioned B. fontanesii and related synonyms and its phytopharmacological activities were cited, the data have been classified according to the ethnomedical use, the phytochemistry and the biological activities of Talghouda. Previous ethnobotanical studies have reported that Talghouda has been used in many folk medicines, especially in thyroid dysfunction, their essential oils and seeds are used in foods in the form of bread. Compositional analysis in different parts of B. fontanesii shows different classes of compounds, including terpenoids and non-terpenoids, as caryophyllene oxide and caryophyllene, (Z)-farnesene, germacrene B, spathulenol, nonacosane and palmitic acid. Phenolic compounds, fatty acids and alkaloids. Polar and non-polar extracts like essential oils have a wide variety of pharmacological activities, i.e., antioxidant, antifungal, antibacterial, anti-inflammatory, anticancer, enzyme inhibition, antiglycation, phytotoxic activity, hypoglycemic effect, estrogenic effects activities. This review confirms the important ethnobotanical and pharmacological potential of Talghouda.

Keywords: Ethnobotany, Talghouda (Bunium fontanesii), ethnobotanical investigations, thyroid dysfunction

1. Introduction

Medicinal plants have been used in healthcare and for food since time immemorial. Currently, almost 80% of the world's population depend on herbal medicines to answer their basic primary health needs for the management of numerous diseases [1].

Among these plants we find Talghouda (*B. fontanesii* Batt. and related synonyms: *B. incrassatum*, *B. mauritanicum*), *Bunium* (Apiaceae) is a widely distributed medicinal plant in Algeria (Quézel and Santa, 1963) [2]. The genus name derives from the Greek term βούνιον (boúnion) [2], which means fatness due to the typical large shape of its tubers. With about 50 species, the genus *Bunium* is distributed in North

Africa, Asia and Europe [4]. Among 212 arid or sub-arid geophytes, this genus includes 128 synonymous species and 31 unresolved species. In particular, four species of *Bunium* are endemic among seven species found in the Algerian flora [2]. The name of the genus is fixed on *Bunium*, in rare cases the genus *Carum* is cited as an equivalent [5]. Regarding Talghouda, we find B. incrassatum, common in the fields; *B. fantanesii* having as synonym *B. mauritanicum* [6]. The names of species bring out at the beginning B. mauritanicum but these are other names cited in Ouézel and Santa [2].

The database "The plant List" cites *B. mauritanicum* as being a synonym of *B. bulbocastanum*, Nevertheless, Miara et al [7] and Benkhalifa et al. [8] specify that it is rather *B.*

bulbocastanum L wich is named Talghouda. Talghouda or Terghouda or Nut or earth gland plant familiar to rural areas in all regions of the tell in Algeria [9]. Talghouda is a perennial plant (geophytes), common in fields and harvests. It is an herbaceous perennial of 30-70 cm, with port of umbelliferous. The stem is slender, furrowed especially towards the top. Its leaves are alternate, 2-3 times divided in narrow strips of general triangular outline and its fruits approximately 2 times longer than broad, with protruding sides, aromatic.

Underground part is a brownish tuber, generally rounded, 1-2 cm in diameter, brownish outside, white inside [10]. The species of *Bunium* genus are aromatic plants with medicinal properties, their grains as well as their essential oil are often used in food and medicine [11]. The use of Talghouda in herbal medicine is very old and is currently experiencing a resurgence of interest among the public. Talghouda (*B. mauritanicum*) plant is well known for its therapeutic virtues among Algerians.

It is found in several regions of Algeria, especially in the North. Talghouda tubers are traditionally and widely used to relieve bronchitis and thyroid disease; and in the treatment of inflammatory hemorrhoids and as antidiarrheals, they are also nutritious and therefore used as food [12]. Talghouda (B. incrassatum), was reported for the first time as medicinal plants in the north Africa and Algeria. It was used to treat asthma, cysts, thyroid disorders and tonsillitis The chemistry of Talghouda [13]. (B.mauritanicum, В. incrassatum, bulbocastanum) has been studied before [12,14-17].

While several researchers have studied the chemical composition of species of the genus Bunium such as: B. hissaricum [18], B. persicum [19-29] B. cylindricum [23], B. alpinum [15], B. brachyactis, B. pinnatifolium, B. sayai and B. microcarpum subsp. Microcarpum [30], B. brachyactis, B. pinnatifolium, B. sayaii and B. microcarpum [31], B. elegans [32,33], B. crassifolium [34]. Algeria has an important biodiversity of Binum species, which constitutes an opportunity for screening of multiple interests like Talghouda (B. fontanesii).

The current review aims to document the ethnomedicinal uses, phytochemistry and biological activities of Talghouda (*B. fontanesii* Batt. and related synonyms). This review is intended to enrich national and global databases of traditional knowledge. To our knowledge, this

study represents the first review performed on Talghouda (B. fontanesii Batt. and related synonyms).

Research methodology

All relevant information about the botanical description, ethnomedicinal uses, phytochemicals and pharmacological activities of Talghouda (B. fontanesii Batt.) were collected from published literature. Therefore, we did not include literature related to molecular, physiological and anatomical aspects. Different sources such as Web of Science, Medline, Scopus, Science-Direct and Google-Scholars were used to explore the published papers on B. fontanesii Batt.

Furthermore, and in order to collect as much data as possible, the different homotypic and heterotypic synonyms of the taxon were used in the research, in particular: Laserpitium fontanesii bulbocastanum Pers., В. auct., Bunium bulbocastanum var. bulbocastanum auct., Laserpitium peucedanoides Desf., bulbocastanum var. peucedanoides (Desf.) J.M. Monts., Bunium fontanesii var. aphyllum Negro, Bunium fontanesii var. Mayor glaucum, Bunium fontanesii var. litorale Maire, Weiller & Wilczek, Carum mauritanicum Boiss. & Reut., Bunium mauritanicum (Boiss. & Reut.) Batt., Bunium fontanesii var. mauritanicum (Boiss. & Reut.) Maire, Bunium perrotii Braun-Blang. & Mayor, Bunium fontanesii var. perrotii (Braun-Blanq. & Maire) Maire, Bunium mauritanicum var. aphyllum Negro, Bunium mauritanicum var. glaucum Maire, Carum retractum Durieu.

In this research, several terms of every synonym of the plant were used: essential oils, antibacterial activity, antifungal activity, antidiabetic activity, Anticancer activity, antioxidant activity.

The work published in French or English mentioning the plant were cited in this review.

The collected manuscripts were identified and examined for relevance based on their titles and abstracts. Reference lists of the retrieved papers were also examined to identify further relevant papers.

Chemical structures were drawn using ChemDraw Pro 8.0 software. PubChem database was used to check the IUPAC names of phytochemicals reported from the plant.

3. Results and discussion

3.1. Botanical description

It is an herbaceous perennial Bulbous plant of 30-70 cm, with an umbelliferous habit, Umbels 5-7 cm wide the stems less thick, and slender, poorly developed; furrowed, especially towards the top, pedicels not indurated, not spreading in a star; the leaves are alternate, 2-3 times divided into narrow strips with a general triangular outline and its fruits about twice as

long as wide, bipinnatisect with linear or linear-lanceolate segments, with prominent sides, aromatic. Underground part generally rounded brownish tuber, 1-2 cm in diameter, brownish outside, white inside [10]. White flowers. fruit of 3-4 mm, without rostrum, with fine primary ribs, well-marked at least on the dry side, with mericarps remaining contiguous to the mattness, 4-5 times longer than wide, reaching 4-5 mm; calyx teeth inconspicuous. Flowering: March-July.



Fig.1. Talghouda (B. fontanesii)

(https://www.flickr.com/photos/20945389@N00/156565744) (https://www.aujardin.info/plantes/*Bunium*-bulbocastanum.php)

3.2. Taxonomy and geographic distribution

3.2.1. Taxonomy and nomenclature

According to Trabut and Marès [35], talghouda is claimed to be *B. incrassatum* and *B. Mauritanicum*, this umbellifer very common in the harvests of the Tell, has a bulky starchy tuber which the natives harvest in years of scarcity. The dried and lightly roasted tubers give an edible flour.

Fresh, the tuber contains an essential acrid product causing intestinal and nervous disorders. In the mountains, B. Alpinum and B. Macuca giving a pleasant tasting tuber. In the oldest flora of Algeria, Battandier et al. [5] give B. mauritanicum as a synonym of Carum mauritanicum Boiss. and Reut. for Algeria (Bousmail, and Atlas high plateaus). Subsequently, Quézel and Santa [2] rather retain B. fontanesii (Pers.) Maire as endemic to North Commonly a spontaneous uncultivated plant except in trials such as those of its introduction in France for a test of adaptation in mountainous regions. This evokes a very interesting case of domestication in progress! If the genus name is fixed on *Bunium*, in rare cases the genus Carum is cited as an equivalent. The names of species bring out at the beginning *B. mauritanicum* but these are other names cited in Quézel and Santa [2].

We find *B. incrassatum*, common in the fields; *B. fantanesii* having as syn. B. mauritanicum, here and there in the fields; *B. chaberti*, endemic to Lalla Khedidja in Djurdjura; *B. elatum* very rare and endemic to Bibans; *B. crassifolium*, also very rare and endemic to El-Kala; B. macua, very rare in Zaccar and Bou Maâd, and *B. alpinum* under the cedars of the Tell Atlas (Algiers, Kabylia and Aurès). Other names are affiliated and accepted, these are *B. atlanticum* (syn. *B. alpium*) and *B. macuca*. Dobignard et al. [36] in (African Plant DB) confirm the maintenance of these taxa. with the addition of others such as *B. carvi*, *B. pachypodium* also existing in Algeria.

The database "The plant list" cites *B. mauritanicum* as being a synonym of *B. bulbocastanum* L. *Bunium* taxa are attached to the Apiaceae family with an umbelliferous inflorescence. This attachment to the Apiaceae

family must evoke the toxic nature even if, for the moment, the aspect of the toxic molecules is not well elucidated [8]. More recently, Dobignard et al. [36] retained B. fontanesii for Algeria, Morocco, Tunisia and Libya.

3.2.2. Geographic distribution

B. mauritanicum Batt. (Apiaceae, Apiales, Magnoliospida) occurs spontaniously in Europe: It can be found in North England [37], Italy and Sicilia [38], Spain [39], Balkan Peninsula, and in

the Mediterranean region where it is very commun [40]. In North Africa, *B mauritanicum* grows natively in Morocco [37]. It can be found in Easthern Algeria especially in Oum Elbouaghi where it is very common [41]. In Tunisia, known as « telghoudi » [42,43], it is considered as a misery food [44]. *B. mauritanicum* grows on several habitats: in clearings and edges of Mediterranean scrublands and brushlands, on chalky, clayey-limestone and rocky soils [45]. It is fairly common on ruderal fields [2].





Fig.1. Bunium fontanesii tubers (¹ Fresh tuber: https://inpn.mnhn.fr/espece/cd_nom/86983/tab/fiche) (²Cut and dried tubers: Photo DAOUD N., 25.11.2021)

3.3. Ethnomedicinal uses

In general, *Bunium* species are herbal plants with healing properties, their essential oils and tubers are used in food and folk medicine all over the world [31,32]. In Algeria, the tubers of this plant are eaten as potato. Dried and powdered, it is considered astringent and antidiarrheal, anti-inflammatory especially for hemorrhoids, and a treatment for bronchitis and cough [12].

In Algeria, B. fontanesii is widely known under the vernacular name of Talghouda. Halimi [46] reports that the flour of this plant is used in medicinal preparations, which is a mildly toxic plant. The plant is known as a laxative, digestive, eliminate flatulence and diuretic. It is also used against angina, stomach aches, kidney stones and tumors. During difficult years, people make bread that is eaten with butter.

The bread of the plant can cause moderate hypnosis, and if it were eaten raw without a veil of fat, the tongue would swell, and the throat would be rough, and if the tumors which were in the legs were bandaged with it, at night they would dissolve. And it was very beneficial. The effects of eating Talghouda are sleepy and numb, and drinking milk fixes them.

In the Mascara region, Benarba et al. [47] states that the plant is used to treat Respiratory tract diseases. Nevertheless, Miara et al [7] report that Talghouda (B. bulbocastanum) is used against flatulence and intestinal worms. Indeed, this phenomenon of one vernacular name attributed to several species of close plants is very common in North Africa, particularly in Algeria, where a single vernacular name such as "Zaater" is given for a sevral species of *Thymus* [48].

Ethnobotanical investigations of *B. mauritanicum* have proposed their potential applications in many disorders, the most advanced use in Algeria is that of the treatment of hypothyroidism by the oral route [9]. The harvested tubers must be dried and then reduced

to flour by means of a mill, the consumption of this flour either in the form of galette, or fresh. However, Djahafi et al. [13] mentionnes that B. *incrassatum* have not been previously reported as medicinal plants in the North Africa and Algeria.

As a therapeutic property the plant has an emollient property. This character marks that Talghouda not only as a food but also as a source of this care. Elsewhere, the seeds constitute a substitute for cumin and also give an oil evoked in traditional treatments.

This plant is also used or the treatment of bronchitis and cough. In the native system of medicine, the dried and powdered tubers are considered antidiarrheal and astringent and have found useful against inflammatory hemorrhoids. They used talghouda flour in medicinal preparations and they are a small poisonous plant. It eliminates bloating and diuresis treats constipation and swelling (Seeds). Dugast [49] analyzed a sample taken from the heights of Arbâa (Blida) and presented the composition in the following %: Water: 15.66; Ashes: 5.50; Nitrogenous matter: 7.00; Fatty matter: 1.34; Starch and congeners: 63.12; Cellulose: 6.40; Undosed materials: 0.98. He drew the conclusion of the resemblance to barley.

By comparing it to the potato, Talghouda seems less rich in starch but better for the rate of fat and nitrogenous matter. He concludes that Talghouda is food. The tuber being the organ harvested in the mountains or in the fields. It can only be eaten when dried and pounded into flour. The dough is then cooked as a mash or in a thicker form to make patties.

By analogy, the tuber is often presented by amateurs as being or resembling Jerusalem artichoke by the shape of its tuber and its turnip taste. On the other hand, Talghouda flour, and after cooking, recalls the taste of barley but probably more refined by its relatively higher level of amino acids. Consumption is specified around the months of February-March, once the

supplies are exhausted and the harvest is not yet ready for beans, peas and Mermez (Barley) [8].

4. Phytochemistry

The chemical compounds of the Talghouda have not been studied enough, there was a vew phytochemical characterization data for of Talghouda extracts and oils like Bousetla et al. [14] on B. incrassatum fruits essential oil from Algeria and Bousetla et al. [12] on B. incrassatum choloromethane methanol roots extract from Algeria, Hayet et al. [15] on B. incrassatum aerial parts essential oils from Algeria, Karouche et al. [16] on B. mauritanicum tubers methanol and aqueous extract and Khadidja et al. [17] on B. incrassatum acetone and hexane extracts from Algeria.

But several studies were on essential oil composition and extract from some Bunium like: Appendino et al. [18] on B. hissaricum petroleum ether extract from Central Asia, Khalid et al. [23] on B. cylindricum seed oil Hexane extract from Pakistan, B. persicum from Iran by Rakhimov et al. [19], Azizi et al. [22] and Jalilzadeh-Amin et al. [25] and Sanei-Dehkordi et al. [27] on essential oil and methanolic extract, Hayet et al. [15] on *B. alpinum* aerial parts essential oils from Algeria, Talebi et al [28] on B. persicum fruits Essential oil, Sharafati Chaleshtori et al. [30] on B. brachvactis (Post) Wolff, B. pinnatifolium Kljuykov, B. sayai Yıld and B. microcarpum subsp. microcarpum (Boiss.) Freyn aerial parts including flowers methanolic extracts from Turkey.

Zengin et al. [31] on methanolic extract, Majidi et al. [29] on *B. persicum* (Boiss.) and *B. Fedtsch* essential oil and methanolic extract, Souilah et al. [34] on *B. crassifolium methanolic extract from Algeria*.

 Table 1. Ethnomedicinal use of Talghouda and most important species of the Bunium genus

Species	Used part	Mode of preparation	Traditional use	References
B. incrassatum Talghouda Algeria	Amo/roots		In local Algerian cookery	[12]
	Amo/tubers		Astringent against cough, bronchitis, diarrhea and hemorrhoids	
Talghouda B. mauritanicum Batt. Algeria	Bulbes		Cough	[50]
B. incrassatum Talghouda Algeria	tubers		Galactagogues to improve breast milk production in the farm animals. Against inflammatory hemorrhoids, bronchitis and cough.	[51]
Talghouda B. mauritanicum Algeria	Whole plant	Trituration	Flu	[52]
Talghouda Algeria B. mauritanicum L.	Seeds	Decoction Raw Maceration Oral SMSD: bones pain	Early-stage cancer Stomach ache	[53]
		Raw Topical	Anxiety disorders and hypochondria	
Talghouda B. incrassatum (Boiss.) Batt. &Trab., Pig Nut Algeria	Tuber	Powder	Stomach ache	[54]
Talghouda B. incrassatum Amo Algeria	Tubers	Infusion, powder Oral	Allergy, asthma, cough, cysts, tonsillitis, thyroid disorders.	[13]
B. persicum (Boiss). B. Central Asia	Fedtsch/fruits	1	To season dishes before the preparation of meat-based foods	[55]
B. persicum (Boiss). Iran	Fedtsch/ whole plant		Gastrointestinal disorders involving indigestion, stomach ache, diarrhea and headache, urinary and respiratory tract infections and colic, diuretic, flatulent, antidiabetic, antiepileptic, antiseptic, antiparasitic, antispasmodic, anticonvulsant and antiasthma remedy, regulate liver function and body weight, increase the milk of lactating mothers	[56,57,58]
			Relieve terrible pains after delivery	[59]
			A parasite repellent	[57]
			Against insomnia, Parkinson, nausea, constipation, convulsion, inflammatory bowel, the blood lipids and cholesterol	[60]
B. bulbocastanum Morocco			Treatment of musculoskeletal and gynecological malfunctions	[61]
B. persicum (Boiss). Iran	Fedtsch/seeds		Stimulant, toxic, address stomach and intestine problems with expectorant, carminative, emmenagogue and galactogogue properties, toothache, jaundice, epilepsy, diarrhea and dyspepsia, appetizer	[56]
			as a spice, condiment and additive to foods and beverages	[62]
B. persicum (Boiss). B. Iran	Fedtsch/fruits		Treat flatulence, dyspepsia, indigestion, colic and dysmenorrhea; anticonvulsant, diuretic, analgesic, anthelmintic and anti-asthma agent	[63,64]

Essential oils

The extraction of essential oils of the genus *Bunium* leads to a wide range of compounds belonging to different classes such as terpenoids and non-terpenoids (Table 1 and Fig.1). The study conducted by Hayet et al. [15] on essential oils composition of Talghouda (*B. incrassatum*), revelated that the main component was palmitic acid. Baser et al. [20] found that p-mentha-1,3-dien-7-al, γ -terpinene, β - pinene, cuminaldehyde and p-mentha-1,4-dien-7-al they the major components of this cumin- smelling B. persicum fruits. Jassbi et al. [32] in another study identified germacrene D and β -caryophyllene as main components for B. elegans oil.

The oil of B. caroides contained α -pinene and (Z)- β -ocimene as major constituents. Previously, Shahsavari et al. [21] found caryophyllene, γ -terpineneand cuminyl acetate as major components for B. persicum essential oil.

The study of Hayet et al. (2017) [15] on essential oils composition of B. alpinum, revelated that caryophyllene oxide was the main component for B. alpinum oil.

Some other species of *Bunium* (*B. pinnatifolium*, *B. brachyactis*, *B. sayaii*, *B. microcarpum*, *B. crassifolium* and *B. persicum*) have been analyzed with LC-MS/MS and showed the presence of phenolic compounds like: Quinic acid, Pantothenic acid, Syringic acid, Naringenin-6.8-di-C-glucoside, Apigenin-C-hexoside-C-pentoside isomer 1, Apigenin-C-hexoside-C-pentoside isomer 2, Luteolin-O-glucuronide, Apigenin-O-glucuronide, Rutin and Kaempferol [31].

In a recent work, Öztürk et al. [33] found, caryophyllene oxide, myristicin, caryophyllenel II and hexadecanoic acid as major components of essential oil for B. elegans (Fenzl) Freyn var. aerial parts collected from Turkey.

Although β -pinene, P-cymene, γ - terpinene and α -pinene, limonene, Z- β -ocimene and E-caryophyllene were the dominant monoterpenes observed predominantly in the fruits oil of B. paucifolium, B. persicum, B. wolffii and B. paucifolium grown in Iran [65].

Phenolic compounds

Regarding Talghouda, few studies were performed and revelated that: The roots of Talghouda (B. incrassatum) from Algeria showed the presence of the oleic acid, β -sitosterol, scopoletin, scoparone and sucrose and β -Sitosterol [12]. The study of Ben Sonia & Zouina [66] on four extracts of methanol, aqueous,

acetone and hexane for *B. incrassatum* roots showed that the highest amount of polyphenols in acetone extract and the hexane and acetone extracts showed the highest levels of flavonoids and also of tannins.

The results of Hayet et al. [15] study showed a highly Total phenolic compounds for Talghouda (*B. incrassatum*) aerial parts methanolic extracts. The acetone extract of Talghouda (B. incrassatum) from M'sila (Algeria) was rich in polyphenols, while the hexane fractions contained the highest amounts of flavonoids, tannins were more frequent in the acetone fraction [17].

Other hands, Talghouda (*B. mauritanicum*) have been studied by the team of Karouche et al. [16] to showed only the chemical screening such as the presence of sterols, triterpenes, saponins, tannins, alkaloids and aglycone flavones. to date, some *Bunium* species have been studied.

Only, we found six species of *Bunium* have been studied with LC-MS/MS and showed the presence of total number of phenolic compounds between 71 and 3 components (Table 1).

We found several classes of natural compounds such as [18] sesquiter- penes [67]. species are: B. pinnatifolium, These brachyactis, B. sayaii, B. microcarpum, B. crassifolium and B. persicum. And the main of components are: Quinic acid, Pantothenic acid, Syringic acid, Naringenin-6.8-di-C-glucoside, hexoside-C-pentoside isomer1, Apigenin-C Apigenin-C-hexoside-C-pentoside isomer Luteolin-O-glucuronide Apigenin-Oglucuronide, Rutin and Kaempferol (Table 1 and Fig.2).

 Table 2. Phytochemistry of Talghouda (B. fontanesii and related synonyms: B. incrassatum, B. mauritanicum, B. bulbocastanum) and somes species of

Runium

				Bunium
Species & country	Used part	Chemical composition / Main compounds	Extract / Method	References
Talghouda B. incrassatum Algeria		oil from fruit: caryophyllene oxide, (Z)farnesene, -caryophyllene, and germacrene B as the principal constituents. oil from fruit-bearing branches: caryophyllene oxide, nonacosane, germacrene B, -caryophyllene, (Z)farnesene, caryophyllenol II, and spathulenol as the principal constituents. Thickened branches: nonacosane, spathulenol, eudesm-4,7-dien-1-ol, caryophyllenol II, (Z)farnesene, germacrene B, and -caryophyllene as the principal constituents.	fruits essential oil	[14]
Talghouda B. incrassatum Algeria	Roots	Oleic acid, β -sitosterol, scopoletin, scoparone, sucrose and β -Sitosterol	Choloromethane- methanol / Mass Spectroscopy (MS) and Nuclear Magnetic Resonance Spectroscopy (NMR)	[12]
Talghouda B. incrassatum Algeria	Aerial parts	The main component was palmitic acid	Essential oils GC/MS	[15]
Talghouda B. mauritanicum Algeria	tubers	presence of sterols, triterpenes, saponins, tannins, alkaloids and aglycone flavones.	methanol and aqueous extract	[16]
Talghouda (B. incrassatum) Algeria		Contain the highest amounts of flavonoids, tannins	Acetone and hexane	[17]
B. brachyactis (Post) Wolff Turkey	Aerial parts including flowers)	Quinic acid, chlorogenic acid,pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin	Methanol extracts / HPLC-MS/MS analysis	[30]
B. pinnatifolium Kljuykov Turkey	Aerial parts/ flowers	Quinic acid, chlorogenic acid,pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin	Methanol extracts HPLC-MS/MS analysis	[30]
B. sayai Yıld Turkey	Aerial parts/ flowers	Quinic acid, chlorogenic acid,pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin	Methanol extracts HPLC-MS/MS analysis	[30]
B.microcarpum subsp. microcarpum (Boiss.) Freyn Turkey	Aerial parts/ flowers	Quinic acid, chlorogenic acid,pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin	Methanol extracts HPLC-MS/MS analysis	[30]
B. pinnatifolium Turkey	Aerial parts	Quinic acid,chlorogenic acid, pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin, B, vicenin-2, Naringenin, kaempferol, Afzelin,	Methanol extract / HPLC-MS/MS	[31]
B. brachyactis Turkey	Aerial parts	Quinic acid, chlorogenic acid, pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin. Vitexin, cosmosiin, diosmin, luteolin, angelicin, salcolin B, vicenin-2, Naringenin, orientin,	Methanol extract / HPLC coupled (HPLC-MS/MS)	[31]

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B. sayaii Turkey	Aerial parts	Quinic acid, chlorogenic acid, pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin, Vitexin, cosmosiin, diosmin, luteolin, angelicin, salcolin B, vicenin-2, Naringenin, kaempferol, Afzelin,	Methanol extract / HPLC-MS/MS	[31]
B. microcarpum Aerial parts Turkey		Quinic acid, chlorogenic acid, pantothenic acid, esculin, isoquercitrin, rutin, apigenin, scopoletin, Vitexin, cosmosiin, diosmin, luteolin, angelicin, salcolin B, vicenin-2, B, vicenin-2, Naringenin, Naringenin, kaempferol, orientin	Methanol extract / HPLC-MS/MS	[31]
B. persicum	Seed (EO)	Essential oil (EO): γ-terpinene (0.8-46.1%), cuminaldehyde (5.96-40.66%),	Methanol extract/ HPLC-MS	[19,25,27,29]
B. Fedtsch Iran	Fruit (FA, CH and PC	α-terpinene-7-al (0.4-37.2%), caryophyllene (0.08-27.81%), γ-terpinene-7-al (8.3-26.91%), ρ-cymene (2.8-19.15%), limonene (0.5-15.7%), β-pinene (0.2-15.62%).	CH/ PC/ Essential oil /GC and GC/MS	
		Carbohydrates (CH): monosaccharides and oligosaccharides (glucose, fructose, mannitol, sucrose, and raffinose), water-soluble polysaccharides, pectin substances, and hemicellulose. Fatty acids (FA): linoleic acid, octadecanoic acid, palmitic acid, petroselinic acid and 8, 11, 14-eicosatrienoic acid. Phenolic compounds (PC): Caffeic acid, ρ-coumaric acid and kaempferol		
B.persicum	fruit	The main constituents were g-terpinene,	Essential oil	[22]
Iran		cuminaldehyde, g-terpinen-7-al	GC and GC/MS	
B. persicum Iran	fruits	The major compounds: γ -terpinene, cuminic alcohol, cumin aldehyde, p-cymene, safranal and limonene	Essential oil GC-FID and GC/MS	[28]
B. crassifolium Algeria	Aerial parts	Chlorogenic, gallic, ferulic acids were the most abundant phenolic acids detected, rhoifolin, quercitrin and rutin were the most abundant flavonoids, fumaric acids, quinic and malic as a non-phenolic organic acid	Methanol extract (70% and 100%) / HPLC-MS/MS	[34]
B. hissaricum Central Asia	Seed oil	Capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, oleic acid, linoleic acid, petroselinic acid, octadec-7enoic acid, octadec-8-enoic acid.	Petroleum ether / Gas-Liquid Chromatography (GLC)	[18]
B. cylindricum Pakistan	Seed oil	Capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, petroselinic acid, oleic acid, linoleic acid, linolenic acid	Hexane / Soxhlet Extraction and Gas- Liquid Chromatography (GLC)	[23]
B. incrassatum		Oleic acid, β-sitosterol, scopoletin, scoparone, sucrose		
B. alpinum Algeria	Aerial parts	The main component caryophyllene oxide	Essential oils GC/MS	[15]
B. cylindricum B. paucifolium B. persicum B. wolffü Iran	Fruit (EO)	The main component of oil: myristicin, β -pinene, α -pinene, apiole, β - selinene, E-caryophyllene, α -selinene, γ -terpinene, γ -terpinene-7-al, cumin aldehyde, myristicin, β -pinene, Z - β -ocimene	Essential oils GC/MS-GC-FID	[65]
B. persicum (Boiss Iran)	Aerial parts	Essential Oil: The major components were caryophyllene (27.81%), γ-terpinene (15.19%), cuminyl acetate (14.67%).	Essential oils/ Hydro-distillation/ Gas Chromatography coupled with FID	[21]
B. persicum (Boiss	Seed Oil	Essential Oil (EO): The major components were cuminaldehyde (21.23%),	Hydro-distillation and GC/MS	[68]

India)		sabinene (14.66%), and γ-terpinen (12.49%). C-Sources // Chemical compounds: D-Arabinose, L-Arabinose, Dextrin, D-	equipped with FID	
		Fructose, L-Fucose, D-Galactose, D-Galacturonic acid, a-D-Glucose, Glucuronamide, D-Glucuronic acid, 2-Keto-D-Gluconic acid, DMannose, Palatinose, D-Psicose, L-Rhamnose, D-Ribose, D-Tagatose, D-Xylose, c-		
		Hydroxy-butyric acid, L-Alanine, L-AlanylGlycine, L-Asparagine, L-Ornithine		
B. persicum (Boiss.) India	Seed Oil	Essential Oil (EO): carvone, Cumin aldehyde, γ-terpinene, α-terpinen-7-al, γ-terpinen-7-al, p-cymene, Limonene, α-pinene, β-pinene, Anisole, Apiole, Carvacrol, Caryophyllene, Cuminic alcohol, Germacrene D, Transdihydrocarvone, α-methylebenzenemethanol,	-	[69]
B. persicum Iran	Seed Oil	Essential Oil (EO): The main constituents were γ-terpinene (24.02%), cuminaldehyde (20.1%), paracymene (13.09%), benzenemethanol alphapropyl (13.01%), cyclopentane, 2-methyl-1-methylene-3-(1-methylethenyl) (3.57%).	Hydro-distillation and GC/MS equipped with FID	[70]
B.kuhitangi B. microcarpum Iran	Aerial parts	Essential Oil (EO): Main components from B. kuhitangi were 9-epi-(E)-caryophyllene (35.38%), α - copaene (8.38%) and δ - selinene (7.35%). * Main components for B. microcarpum. were 9-epi-(E)- caryophyllene (73.61%), γ - cuprenene (8.37%) and α -cadinene (5.75%).	Hydro-distillation and GC/MS	[71]
B. Persicum (Boiss) Iran	Seed	Essential Oil: Tricyclene, α -thujene, sabinene, β -pinene, myrcene, p-cymene, Limonene, γ -Terpinene, linalool, terpioneole, cuminaldehyde, α -terpinene-7-al, γ -terpinene-7-al,	Hydro-distillation (HD), ultrasound- assisted extraction (UAE with nhexane) and Soxhlet extraction (SOX with n- hexane) and GC/MS equipped with FID	[72]
B. persicum B. Fedtsch. Iran	Seed	Major essential oil γ-terpinene, cuminaldehyde	Methanol with percolation method and Hydro-distillation	[73]
B. persicum (Boiss.) Fedtch Iran	Seed	Essential Oil: the γ-Terpinene (46.1%) and cuminaldehyde (15.5%)	Petroleum ether, chloroform, methanol and water with percolation method, to the essential oil isolated using water-distillation method/ GC/MS-FID	[74]
B. persicum B. Fedtsh Iran	Fruits	Essential Oil: 1,4-p-menthadien-7-al, p-menyha-1,3-dien-7-al, cuminal, trans-isocarveol, γ-terpinene, limonene, p-cymene, β-pinene, sabinene, α-pinene	Ethanol, Hydro-distillation, GC/MS	[63]
B. persicum Boiss. Iran	Seed	Volatile compounds: α -Thujene, α -Pinene, Sabinene, β —Pinene, Myrcene, ρ -Cymene, Limonene, γ —Terpinene, Terpinen-4-al, Cumin aldehyde, α -Terpinen-7-al, γ -Terpinen-7-al	Superheated water extraction (SWE), Hydrodistillations, Soxhlet extractions and GC/MS and GC-FID	[75]
B. persicum Bois s Iran	Seed	* γ -terpinene(37.98%), cuminaldehyde (11.48%) and α -methylbenzenemethanol (25.55%).* ρ -cymene in the hydrodistilled essential oil, also γ -terpinene and cuminaldehyde	Supercritical fluid extraction (SFE), Hydrodistillations and GC/MS	[76]

Fatty acids

The main compounds of fatty acids are capric acid, lauric acid, myristic acid, palmitic acid and stearic acid (**Table 1 and Fig.3**) were found in the seed oil of B. hissaricum Korovin and B. cylindricum (Boiss. & Hohen.) Drude by GLC (Appendino et al., [18] and Khalid et al. [23].

Carbohydrates

The main compounds of carbohydrates are monosaccharides and oligosaccharides (glucose, fructose, mannitol, sucrose, and raffinose) (**Table 1 and Fig.4**), water-soluble polysaccharides, pectin substances, and hemicellulose were found in B. persicum seed oil [19,29].

Fig.1. Chemical structure of the main compounds of essential oil of some *Bunium* species *studied*

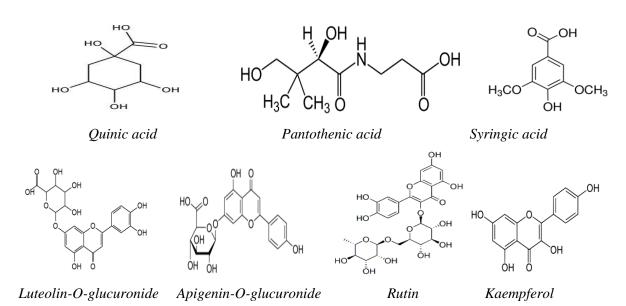


Fig.2. Chemical structures of the main phenolic compounds of some Bunium species studied

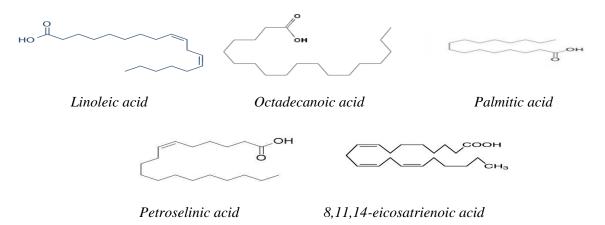


Fig.3. Chemical structures of the fatty acids of B. persicum

Fig.4. Chemical structures of mono and oligosaccharides of B. persicum

3.6. Biological activity

The extracts derived from Talghouda and other species of *Bunium* species are known to possess remarkable biological activities, which are discussed in the following subsections. Not all the biological properties have been studied and not all the *Bunium* species have been tested so far.

3.6.1. The antioxidant activity

The total capacity of antioxidants to scavenge the free radicals, via their hydroxyl substituents and their hydrogen donating ability may be defined as the antioxidant activity. This mechanism facilitates antioxidants to protect DNA or cell walls against the oxidative stress [77]. Therefore, it is associated with many other biological activities including antimutagenicity, anticarcinogenic and antiaging [65].

The antioxidant activity is mostly contributed to the secondary metabolites of the plants, as part of their defense mechanism. The antioxidant activity was depicted in Talghouda (Table 3). The research by Karouche et al. [16] revealed that the methanolic extract of B. mauritanicum tubers had a higher amount of total phenolics 4.031 mg QE/g of extract, although the water extract was had a higher antioxidant activity (IC50 of 0.14 mg/ml against the DPPH radical) than the methanolic extract.

Moreover, results of Hazarika and Das [78] suggests that all fractions of B. bulbocastanum

extract (aqueous, crude methanolic, ethyl acetate, n-hexane and chloroform) had antioxidation potential with an encouraging finding of percent antioxidation inhibition.

Otherwise, Ahmad et al. [79] indicated that Ethyl acetate, aqueous, crude methanolic, n-hexane fraction of B. bulbocastanum fruits have an antioxidation potential. However, methanolic extracts of B. incrassatum and B. alpinum from Algeria exhibited a highest antioxidant activity, essential oil which gave the lowest values [15].

The same finding by Lefahal et al. [80] who evaluated the antioxidant activity of ethyl acetate and n-Butanol extracts and four flavonoids that had been isolated from the aerial parts of B. alpinum.

The n-Butanol fraction showed the best antioxidant activity. In contrast, the isolates demonstrated varying degrees of antioxidant activity. B. incrassatum from M'sila (Algeria) was also studied by Khadidja et al. [17] to evaluate the antioxidant abilities of the water, methanol, acetone and hexane fractions, the best scavenging activity was obtained by hexane followed by acetone and methanol fractions, acetone fractions showed the best iron reducing activity.

In other investigation, ethanol and ethyl acetate extracts of B. bulbocastanum were evaluated in term of total phenolic and flavonoid content of the seeds, highest TPC was observed

in B. bulbocastanum ethyl acetate extract while the lowest was observed in ethanol extract.

The seed essential oils of B. persicum Boiss from Iran were also revealed to have the antioxidant properties, with their relatively lower antioxidant activity (IC50 of 0.88 mg/mL against the DPPH radical) [21]. Among the 4 different methanolic extracts of B. species (B. brachyactis (Post) Wolff; B. pinnatifolium Kljuykov; B. sayai Yıld and B. microcarpum subsp. microcarpum (Boiss.) Freyn) from Turkey, the highest antioxidant activity was detected for *B. pinnatifolium and B. microcarpum* by most of the six assays used in scope of the study [31].

According to the research group, the relatively higher antioxidant activity results from these two species are mainly due to their higher total phenolic and flavonoid contents, as the highest amount of total phenolics was observed in the species B. pinnatifolium (35.94 mg GAE g-1) and the highest total flavonoids were detected in B. microcarpum and B. pinnatifolium, respectively (39.21 mg RE g-1 and (38.33 mg RE g-1) [31].

Although the differences among the total phenolic and total tannin contents of different parts (corm, leaf, stem and fruit) were not significant (p>0.05) for the 4 different *Bunium* species of Iran, the researchers revealed that the total phenolic content of the leaves and stems of *B. paucifolium* (6.10 and 6.03 mg GAE/g DW, respectively) and leaves of B. wolffii (6.07 mg GAE/g DW) were significantly higher than the

others other 2 species *B. Cylindricum* (Boiss. & Hohen.) Drude and *B. persicum* (Boiss.) [65].

According to the same study, The IC50 values of the different parts of these 4 species against the DPPH radical changed from 27.15 to 71.31 mg/mL. The highest antioxidant activity results were measured in the methanolic extracts (leaves, fruits and stems) of species *B. persicum* (27.15, 27.33 and 27.76 mg/mL, respectively) [65].

These results may be considered as encouraging for the potential of unidentified species of the *Bunium* species, too. The antioxidant activity of the essential oil from certain *Bunium* species (*B. persicum Boiss.*) has also been determined.

According to the findings, the essential oil had a promising antioxidant activity (IC50 value of 15.12 μ L/mL against the DPPH radical). Furthermore, nanoencapsulated *B. persicum Boiss*. essential oil (in a chitosan-cinnamic acid based nanoemulsion system) has a slightly lower IC50 value against the DPPH radical activity (12.64 μ L/mL) in comparison to its free counterpart [68].

3.6.2. The antimicrobial activity

Talghouda tubers are endowed with antibacterial properties (Table 4), they can act against Gram+ Staphylococcus aureus and Gram- Pseudomonas aeroginus [16].

Table 3. Antioxidant activity of Talghouda (B. fontanesii and related synonyms: B. incrassatum, B. mauritanicum, B. bulbocastanum) and species of Bunium

Species	Part used	Extract	Methods	Keys results	Ref.
Talghouda B. bulbocastanum	Fruits	Ethyl acetate, aqueous, crude methanolic, n-hexane fraction	*Antioxidation potential (Nitric Oxide (NO) free radical scavenging assay)	Ethyl acetate, aqueous, crude methanolic, n-hexane fraction has an antioxidation potential	[79]
Talghouda B. Bulbocastanum Kashmir/India	Fruits	Ethyl acetate, Aqueous, Crude methanolic, n- Hexane and Chloroform fractions	Nitric Oxide Method	*Ethyl acetate fraction had a potent antioxidation (30.34%) at 0.5 mg/ml *_Aqueous fraction showed significant antioxidation potential of 26.07%. *The rest of the three fractions had a lower antioxidation activity.	[78]
Talghouda B. incrassatum Algeria	Aerial parts	*Methanolic extracts *Essential oils	Antioxidant activity DPPH technique	Methanolic extracts gave highest antioxidant activity compared to the Methanolic extracts	[15]
Talghouda B. mauritanicum Algeria	tubers	Methanolic extract	*Total phenolics content * Antioxidant activity (DPPH)	*4.031 mg QE/g of extract * IC50 of 0.14 mg/ml	[16]
Talghouda B. incrassatum Algeria		Methanol, acetone and hexane extracts	Antioxidant abilities	*The best activity by hexane extract followed by acetone and methanol extracts, *Acetone fractions showed the best iron reducing activity.	[17]
B. persicum Boiss Iran	Seed	Essential oils	Antioxidant properties DPPH	lower antioxidant activity IC50 of 0.88 mg/mL	[21]
B. alpinum_Algeria	Aerial parts	methanolic extracts Essential oils	antioxidant activity DPPH technique	Methanolic extracts gave highest antioxidant activity compared to the Methanolic extracts	[15]
B. alpinum	Aerial parts	* Ethyl acetate and n- Butanol extracts *flavonoids that had been isolated	*Antioxidant activity	*n-Butanol extract showed the best antioxidant activity against ethyl acetate	[80]
B. brachyactis B.pinnatifolium B. sayai B.microcarpum subsp. microcarpum Turkey	Aerial parts	Methanolic extracts	* Total phenolic contents *Antioxidant properties DPPH, ABTS, CUPRAC, and FRAP assays	*Highest antioxidant activity for B. pinnatifolium and <i>B. microcarpum</i> *Higher total phenolic contents in B. pinnatifolium (35.94 mg GAE g-1) *Highest total flavonoids in B. microcarpum and B. pinnatifolium, respectively (39.21 mg RE g-1 and (38.33 mg RE g-1)	[31]
Bunium species B.persicum B. persicum		Essential oil	*Antioxidant activity DPPH	 * Essential oil had a promising antioxidant activity (IC50 value of 15.12 μL/mL * Lower IC50 value against the DPPH radical activity (12.64 μL/mL) 	[68]
B. paucifolium B. wolffii B. cylindricum B. persicum Iran	Corm, Leaf, Stem and Fruit	Methanolic extracts	*Total phenolic and total tannin contents *_Antioxidant activity DPPH radical	*Total phenolic content of leaves and stems of <i>B. paucifolium</i> (6.10 and 6.03 mg GAE/g DW, respectively) * Leaves of <i>B. wolffii</i> (6.07 mg GAE/g DW) higher than the others 2 species <i>B. Cylindricum</i> and <i>B. persicum</i> * IC50 values of 4 species changed from 27.15 to 71.31 mg/mL.	[65]
B. crassifolium Algeria		Methanol and methanol water (70:30) extracts.	*Antioxidant activities: DPPH, ABTS, β-carotene, CUPRAC, Fe+2 assays	*The methanol:water extract showed the high value of antioxidant activities.	[34]

These antimicrobial properties are due to the presence of couramins in its chemical content [12]. Plants have used against the infections for ages. However, the current emergence of infectious diseases and the rise of microbial resistance, revalue them as antimicrobial agents.

According to the findings from the study by Zengin et al. [31], the antimicrobial activity was evident and found as variable among the different *Bunium* species of B. brachyactis (Post) Wolff; B. pinnatifolium Kljuykov; B. sayai Yıld and B. microcarpum subsp. microcarpum (Boiss.) Freyn) from Turkey [31].

The certain bioactive components found in the *Bunium* species, was previously found to possess antimicrobial activity against *P. aeruginosa*, *S. Typhimurium*, and *P. mirabilis* and/or antioxidant activity [81,82].

Therefore, their findings revealed that although these species depicted a wide range of antimicrobial activity against different bacteria in the scope of the study, they were efficient against P. mirabilis and E. coli, mainly [31]. The results for antibacterial study of Khan et al. [83] aimed at screening the B. bulbocastanum for its antibacterial and antifungal, activities.

The methanolic extract and n-hexane fraction showed significant activity against Staphylococcus aureus while, the cloroforme fraction was moderately active against S. aureus. A moderate activity was shown by the ethyl acetate fraction against B. subtilis. This fraction was inactive against P.aerogenosa and S.aureus. The aqueous extract showed significant activity against B. subtilis, and moderate activity against S. aureus and E. coli and low activity against P.aerogenosa. regarding the antifungal assay, the same stydy indicated that all the test extracts were inactive against all the test fungi.

The study of Bousetla et al. [12] reveals that the crude extract (Choloromethane-methanol: 1/1) from B. incrassatumexhibited stronger activity against fungi than bacteria strains. EOs of B. incrassatum and B. alpinum showed wide array of antibacterial activity against bacteria [15].

Particularly the essential oil from some *Bunium*, such as B. persicum depicted also antibacterial activity against E. Coli [84] and antifungal activity against different species of *Fusarium oxysporum* [70], although the difficulties in its food applications have also been mentioned referring to its high volatility, strong

odor, hydrophobic nature, and unknown mode of action.

Therefore, these researchers hypothesized that, the antifungal efficacy of B. persicum essential oil might be increased by encapsulating in a chitosan-cinnamic acid based nanoemulsion system [68]. Fungal contamination is a major food safety issue. It is mainly related with the improper post-harvest storage.

Other activities

From the investigation of Hazarika and Das [78] using MTT assay, we are able to conclude that aqueous and ethyl acetate extract of *B. bulbocastanum* fruit has a noteworthy anticancer activity.

Another study of the enzyme inhibitory test of aerial parts methanol extracts of B. sayai, B. pinnatifolium, B. brachyactis and B. macrocarpum, showed a high activity against cholinesterase, tyrosinase, amylase, glucosidase and lipase [31].

The findings of Ahmad et al. [79] for the antiglycation in vitro assay of the ethyl acetate, aqueous, crude methanolic and n-hexane fraction of B. bulbocastanum fruits showed that, among the tested samples, chloroform fraction was the most effective antiglycation agent with MIC50 of 132.88 μg/ml followed, and the percent inhibition of glycation is concentration dependent.

The finding of khan et al. [83] phytotoxic activity of B. bulbocastanum showed that all the test samples were unable to agglutinate red blood cells of the human blood indicating that this species lack phytolectins. Also, Hammoudi et al. [51], in its study, evaluated the effect of the treatment of pregnant rabbits of B. incrassatum by roots extract, on hematological and histological parameters of the adrenal gland during the last third of pregnancy, results suggests that B. incrassatum had a hypoglycemic effect and is safe for use up to 100 mg/kg/day for both nutritional and medicinal purposes which open new perspectives for this plant in the bioactive materials technology.

In a recent study on B. incrassatum and B. alpinum from Algeria, methanolic extracts were used in studying in vitro anti-inflammatory activity using egg albumin technique and in vitro anti-hemolytic activity using HBRC technique [15] findings exhibited a significant antihemolytic and anti-inflammatory activity [41].

						Бипшт
Species	Part used	Extract	tested/ compound	Tested strains	Keys results	References
Talghouda		Methanolic	Antibacterial	*Staphylococcus aureus ;	* CHCl3 not active against S. aureus	[83]
B. bulbocastanum		extract and n-	activities	B. subtilis ;	* Low activity against B. subtilis (ethyl acetate fraction)	
		Hexane fraction		P. aerogenosa ; E. coli Fungi.	* Moderate activity against Aeruginosa and S. aureus (ethyl acetate fraction)	
					* Aqueous extract showed significant activity against B. subtilis,	
					moderate against S. aureus and E. coli and low activity against P.	
					aerogenosa.	
					* All the test samples were inactive against all fungi.	
Talghouda		Cholorommet	Antimicrobial		*Stronger activity against fungi than bacteria strains	[12]
B. incrassatum		hanol	activities			
Talghouda			Antibacterial		Essential oil showed wide array of antibacterial activity against	[15]
B. incrassatum			activities		Gram-positive and Gram-negative bacteria.	
B. alpinum						
B. persicum		Essential oil	Antifungal activities	Fusarium oxysporum	Antifungal activity against six different species of Fusarium oxysporum	[70]
B. brachyactis	Aerial	Methanol	Antibacterial	Proteus mirabilis	*P. mirabilis active against E. coli (MIC and MBC < 1 mg	[31]
B. pinnatifolium	parts	extracts	activity	E. coli	mL-1).	
B. sayai			Antifungal,	Aspergillus versicolor	*B. brachyactis effective against A. versicolor	
B. microcarpum			activities.	Trichoderma viride	* B. brachyactis effective against Trichoderma viride.	
Turkey				Aspergillus fumigatus	*B. sayai effective in inhibiting A. fumigatus.	
B. persicum		Essential oil	Antibacterial activities	E. Coli	Antibacterial activity against E.Coli	[84]

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					Випіит
Species	Part used	Extract	Methods	Keys results	References
Talghouda <i>B.bulbocastanum</i>	Fruit	Ethyl acetate and aqueous fraction	Anticancer activity using MTT assay	Ethyl acetate and aqueous fraction has a noteworthy activity on human cell lines	[78]
Talghouda B.bulbocastanum		methanolic extract	Phytotoxic activity		[83]
Talghouda B. incrassatum B. alpinum Algeria		Methanolic extracts	In vitro anti-inflammatory activity In vitro anti-hemolytic activity using HBRC technique	significant antihemolytic and anti-inflammatory activity	[15]
Talghouda B. incrassatum			Estrogenic effects	*Decrease in the level of Triglyceride cholesterol *Significant increase in growing of follicles accompanied by decrease in atretic follicles	[41]
Talghouda B. incrassatum	roots		Hypoglycemic effect/ egg albumin technique	Hypoglycemic effect	[51]
Talghouda B. bulbocastanum	Fruits	Ethyl acetate, aqueous,methanolic, n-hexane	Antiglycation in vitro assay	*Chloroform fraction most effective antiglycation agent with MIC50 of 132.88 $\mu g/ml$	[79]
B. sayai, B.pinnatifolium, B. brachyactis B. macrocarpum,	Aerial parts	Methanol extracts	Enzyme inhibitory test	*High enzyme inhibition against cholinesterase, Tyrosinase, amylase, glucosidase, and lipase	[31]
B. crassifolium Algeria		Methanol and methanol water (70:30) extracts.	-Anticholinestrase -Antityrosinase	*The methanol extract showed a good acetylcholinesterase inhibitory, and butyrylcholinesterase inhibitory activity, demonstrated that the both extracts showed a weak activity. -The methanol extract showed a good tyrosinase inhibitory activity more than the methanol:water	[34]

In aim to evaluate the effect of the treatment of mature female rabbits by organic extract of *B. incrassatum* roots, cited that the treatment induces a significant decrease in the level of Triglyceride, Cholesterol and a significant increase in the number of growing follicles accompanied by a decrease in atretic follicles, compared to the control group. The study shows that the organic extract of B. Incrassatum has estrogenic effects (Table 5).

Conclusion and future perspectives

This survey reported review of Talghouda concerning ethnotheir medicinal use, geographic distribution, taxonomy and biological activities. Talghouda is a species complex of the Apiaceae family.

Its starchy tuber is harvested to extract a food flour reminiscent of old eating habits in rural. This tuber is still used in treatments by herbalists. Its interest in the treatment of thyroid dysfunction is also mentioned.

An analysis of biodiversity is needed to shed light on the differences observed between taxa to specify those due to genetics and those due to the effects of the environment. The phytochemical analysis showed the richness of this plant, especially terpenoids in the essential oils which were dominated by caryophyllene oxide and caryophyllene, (Z)--farnesene, germacrene B, spathulenol, nonacosane, alkaloids and fatty acids and carbohydrates as monosaccharides and oligosaccharides.

However, there is a lack of data on flavonoids and phenolic acids in seeds, roots, and fruits. Thus, investigation of these chemical classes in these parts should be explored using different analytical tools such HPLC, GC-MS, and LC-MS. Furthermore, pharmacological effects of Talghouda extracts and essential possessed many biological effects, including antioxidant, antifungal, antibacterial, antiinflammatory, anticancer, enzyme inhibition, antiglycation, phytotoxic activity, hypoglycemic effect, estrogenic effects activities.

However, elaborating the antidiabetic, toxicological and other studies is necessary

to prove these compounds' effects on human health.

Moreover, no literature informations are available on the toxicological of Talghouda. Therefore, to determine the safety of this plant, further toxicological studies are necessary.

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Disclosure Statement

The authors report no conflict of interest.

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