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***Inventory of melliferous plants and physicochemical
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بِسْمِ اللَّهِ الرَّحْمَنِ
الرَّحِيمِ

Dedication

To my Mother Akila Benamour and Father Ahmed

To my Brothers

To my Sisters

To all my Nephews and Nieces

To my whole family

To my friends

To all biology faculties and students

To all who loves science and nature

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Table of contents	
Introduction	1
Bibliographical part	4
Chapter I. Honeybee, melliferous plants and beekeeping	5
1. Honeybee	6
1.1. Classification	6
1.2. Algerian honeybee' subspecies	7
2. Melliferous plant	7
2.1. Introduction	7
2.2. Definition	8
2.3. Nectar	8
2.4. Pollen	9
2.5. Pollination	11
2.5.1. The different modes of pollination	11
2.5.2. Pollination by insects	11
2.5.3. Characteristics of the entomophilous flower	12
2.5.3.1. Factors that ensures the approach and the visit of insects	12
2.5.3.2. The floral characteristics which favor the transfer of pollen by insects	12
2.5.3.3. The arrangement of nectaries in the flowers	12
2.5.4. Variation and condition of the pollen supply	13
2.6. The relationships between the bee and the flower	13
2.6.1. The relationship of the honeybee with the flower	13
2.6.2. The relationship of flowering plants with the honeybee	14
3. Beekeeping	15
3.1 History	15
3.2. Beekeeping advantages	15
3.2.1. Agronomic advantages	15
3.2.2. Economic advantages	15
3.2.3. Therapeutic advantages	16
3.3. The beehive	16
3.4. The Apiary	16

3.5. Beekeeping value of the plant cover	17
3.6. Variation in honey production	17
3.6.1. Intensity of foraging	18
3.6.2. Soil	18
3.6.3. The light	18
3.6.4. The climate	18
3.6.5. The temperature	18
3.6.6. The air humidity	18
3.6.7. The latitude	19
3.7. The floral calendar	19
3.7.1. Definition	19
3.7.2. The stages of making a floral calendar	19
Chapter II. Beekeeping products	20
1. Honey	21
1.1. Chemical composition of honey	21
1.1.1. Sugars	21
1.1.2. Amino acids and proteins	22
1.1.3. Organic acids	24
1.1.4. Phenolic compounds	24
1.1.5. Volatile compounds	25
1.1.6. Vitamins	26
1.1.7. Minerals	27
1.1.8. Pesticides	27
1.2. Physico-chemical properties	28
1.2.1. Moisture	28
1.2.2. Acidity	29
1.2.3. pH	29
1.2.4. Electrical conductivity and ash	29
1.2.5. Hydroxymethylfurfural (5-HMF)	29
1.2.6. Diastase activity	30
1.2.7. The density	30

1.2.8. The viscosity	30
1.2.9. Color	31
1.2.10. The specific heat	31
1.3. Biological properties	31
1.4. Honey of nectar	32
1.4.1. Unifloral honeys (monofloral)	32
1.4.2. Multi-floral honeys or poly-floral honeys	33
1.5. Honeydew honey	33
2. Pollen	33
2.1. Definition	33
2.2. Compositions	34
2.3. Usage	34
2.5. Pollen collection and conservation	35
3. Beeswax	35
3..1. Definition	35
3.2. Compositions and properties	36
3.3. Usage of beeswax	36
4. Propolis	36
4.1. Definition	36
4.2. Compositions and properties of propolis	37
4.3. Usage	37
5. Honeybee venom	37
5.1. Definition	37
5.2. Compositions and properties	37
5.3. Usage	38
6. Royal jelly	38
6.1. Definition	38
6.2. Compositions	38
6.3. Usage	39
Experimental part	40
Chapter I. Inventory of melliferous plants	41

1. Materials and Methods	42
1.1. Study Area	42
1.2. Identification and characterization of the melliferous flora	43
2. Results and discussion	44
2.1. Inventoried melliferous plants	44
2.2. Classes and families of inventoried honey plants	52
2.3. Morphological types	54
2.4. Cultivated flora	56
2.5. Flowering schedule	57
2.6. Beekeeping value of melliferous flora	60
2.7. Abundant and nearby honey plants	61
2.7.1 Abundant and nearby melliferous plants of 6m zone	61
2.7.2. Abundant and nearby melliferous plants of 70m zone	63
2.7.3. Abundant and nearby melliferous plants of 700m zone	64
2.7.4. Similarities between the stations	65
2.7.5. Flowering periods	66
Chapter II. Honey analyzes	67
1. Materials and methods	68
1.1. Samples	68
1.2. Physicochemical analyses	69
1.2.1. Moisture content	69
1.2.2. pH	69
1.2.3. Free, lactonic and total acidities	70
1.2.4. Electrical conductivity (EC)	70
1.2.5. Ash	70
1.2.6. Hydroxymethylfurfural (HMF)	71
1.3. Protein content	71
1.4. Color analysis	71
1.5. Total phenolic contents	72
1.6. Total flavonoid contents	72
1.7. DPPH radical scavenging activity	72

1.8. Reducing power	72
1.9. Antibacterial activity	73
1.10. Pesticides	73
1.10.1. Pesticides analysis	73
1.10.2. Photodegradation of organophosphorus (OPs) pesticides	74
1.11. Statistical analysis	75
2. Results and discussion	75
2.1. Physicochemical parameters of honeys	75
2.1.1. Moisture content	75
2.1.2. pH	77
2.1.3. Free, lactonic and total acidities	78
2.1.4. Electrical conductivity (EC)	81
2.1.5. Ash content	83
2.1.6. Hydroxymethylfurfural content	85
2.2. Protein content	86
2.3. Color analysis	88
2.4. Total phenolic content	89
2.5. Total flavonoid content	91
2.6. DPPH radical scavenging activity	92
2.7. Reducing power	94
2.8. Antibacterial activity	96
2.9. Pesticides	97
2.9.1. Pesticides analysis	97
2.9.2. Organophosphorus pesticides photodegradation	99
Conclusion and perspectives	105
Appendix 1	108
Appendix 2	119
Bibliographical references	127

List of tables

Table 1. Rate of pollen supply of some plants.	13
Table 2. Main honey enzymes and their activities.	23
Table 3. Average contents of free amino acids in honey.	23
Table 4. Phenolic compounds found in honey.	25
Table 5. Pesticide residues reported most frequently in honey.	28
Table 6. Inventory, beekeeping value, period of flowering, morphological types and domestication of melliferious plants of Jijel.	46
Table 7. Classes of melliferous plants.	52
Table 8. Families' diversity of melliferous plants.	53
Table 9. Distribution of morphological types of planted melliferous flora.	57
Table 10. Abundant and nearby melliferous plants of 6m zone.	62
Table 11. Abundant and nearby melliferous plants of 70m zone.	63
Table 12. Abundant and nearby melliferous plants of 700m zone.	64
Table 13. Geographical information of samples.	68
Table 14. Color analysis of tested honeys.	89
Table 15. Pearson correlation coefficients among parameters.	95
Table 16. Antibacterial activity of tested samples.	96
Table 17. Summary table of the results of the analysis of the optically active organophosphorus pesticides by GPC chromatography of the honey samples recollected in different region of Jijel.	98

List of figures

Figure 1. <i>A. mellifera sahariensis</i> (left picture) and <i>A. mellifica intermissa</i> (right picture) entering hives.	07
Figure 2. Honeybee foraging a flower of <i>Taraxacum officinalis</i> .	08
Figure 3. Chemical composition of U.S. honey.	22
Figure 4. Locations of the three inventoried stations in Jijel.	42
Figure 5. Distribution of melliferous plants at the three stations.	45
Figure 6. Morphological types of melliferous flora of Jijel.	55
Figure 7. Distribution of morphological types according to the stations.	56
Figure 8. Number of blooming species during each month in Jijel region (2015-2017).	59
Figure 9. Duration of flowering time of melliferous plants in Jijel region (2015-2017).	59
Figure 10. Distrubution of polliniferous and nectariferous plants.	61
Figure 11. Geographical location of samples.	68
Figure 12. Experimental solar photoreactor support for honey photodegradation reaction.	74
Figure 13. Moisture content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	76
Figure 14. pH of of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	77
Figure 15. Free acidity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	78
Figure 16. Lactonic acidity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	79
Figure 17. Total acidity of honeys from coastal (C1-C11) and mountain (M1-M11 regions of Jijel (Algeria).	80
Figure 18. Electrical conductivity (EC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	82

Figure 19. Ash content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	84
Figure 20. Hydroxymethylfurfural (HMF) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	86
Figure 21. Protein content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	87
Figure 22. Total phenolic content (TPC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	90
Figure 23. Total flavonoid content (TFC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	92
Figure 24. DPPH radicals scavenging activity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	93
Figure 25. Reducing power (RP) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria).	95
Figure 26. Preliminary chromatographic analysis (GC) of honey samples on the HP-608 polysiloxane capillary column.	100
Figure 27. Photodegradation evaluation of OPs pesticides in honey under solar light irradiations a) Beni belaid region, b) El kennar region, c) Achouat region.	101
Figure 28. Pseudo-first order kinetics of photodegradation data of OPs pesticides a) Beni belaid region, b) El kennar region, c) Achouat region.	103

Abbreviation list

ATCC	American Type Culture Collection
DPPH	2,2-diphenyl-1-picrylhydrazyl
EC	Electrical conductivity
ECD	Electron capture detector
FRAP	Ferric reducing antioxidant power
GC	Gas chromatograph
HMF	Hydroxymethylfurfural
IC₅₀	The half maximal inhibitory concentration
LOD	Limit of detection
LOQ	Limits of quantification
MRLs	Maximum Residue Levels
OPs	Organophosphorus
RP	Reducing power
RSA	Radical scavenging activity
TEAC	Trolox equivalent antioxidant capacity
TFC	Total flavonoid contents
TPC	Total phenolic contents

Introduction

Introduction

Honey is a food that humanity has known since the dawn of time. Its uses by the ancient humans are varied (Molan 2001; Khan et al. 2017); it was employed in ophthalmology and ear diseases and played a crucial role in the food and pharmacopoeia to treat burns, snake bites or infected wounds (Cernak et al. 2012). Honey is a natural product that honeybees produce from some plant parts or excretions of some insects that feed on plant sap (Karabagias et al., 2014). More than two hundred components have been found in honey; it is an important source of energy due to its high sugar content, mainly fructose (38%) and glucose (31%) (Alvarez-Suarez et al., 2010; Bueno-Costa et al., 2016). Moreover, it has small amounts of amino acids, proteins, phenolic compounds, carotenoids, organic acids, ascorbic acid, enzymes, α -tocopherol, and oligosaccharides (Alvarez-Suarez et al., 2010). The composition and characteristics of honey are primarily determined by the food source (plants); however, environmental factors, processing, and storage affect this composition as well (Saxena et al., 2010).

To evaluate the characteristics and bioactive properties of honey, phenolic compound content and antioxidant activity have been widely used as indicators (Tahir et al., 2017). Honey contains a variety of phenolics, and it is rich in antioxidants, which increases its usability potential for therapeutic purposes (Küçük et al., 2007). In addition, several others have mentioned the antimicrobial potential of honey (Küçük et al., 2007; Alvarez-Suarez et al, 2010; Liu et al., 2013; Bueno-Costa et al., 2016). The concentration of hydrogen peroxide, which is determined according to the level of glucose oxidase (from bees) and catalase (pollen source), in honey mainly predicts its antimicrobial potential, however, lysozyme, phenolic acids, and flavonoids are the major non-peroxide contributing factors (Tenore et al., 2012). On the other hand, the correlation of the color with bioactive compounds and antioxidant and antibacterial activities has been revealed in other studies (Bueno-Costa et al., 2016). In recent years, many authors have studied the physicochemical and bioactive properties of honeys from different regions in the world including Algeria (Ouchemoukh et al., 2007; Tenore et al., 2012; Bueno-Costa et al., 2016; Mouhoubi-Tafinine et al., 2016; Tahir et al., 2017), using different analytical methods. In addition, Pesticides contamination has been revealed by several researchers in various honey samples collected in different regions of the world (Rissato et al., 2007; López et al. 2014; Abdallah et al. 2017). Most honeys produced in the world are now contaminated by insecticides of the family of neonicotinoids, referred to as "bee killers" (Johnson et al. 2010).

In many flowering plants, the pollination is ensured by insects, the most qualified among them are undoubtedly the bees, which visit the flowers in order to collect nectar and pollen for the preparation of honey (Dumas et al., 1984). Experiments have shown that honeybees pollination double and even triple the production of seeds and fruits (Chauvin, 1968). The plants visited by the honeybee, called melliferous plants, determine the characteristics of honey and their taste (Cherif, 1990). Unfortunately, the heritage of melliferous flora of Jijel like other Algerian regions is not taken into account either by disinterest, lack of means and especially by lack of information.

The aim of this study is to inventory the melliferous flora of Jijel, to evaluate the characteristics (physicochemical properties, protein content, color parameters, contents of total phenolics and total flavonoids, DPPH radical scavenging activity, reducing power, antibacterial activity and pesticides contamination) of honeys from the greenest region of Algeria (Jijel) from different altitudes, to determine the differences between coastal and mountain honeys to reveal the correlation between the altitude and different parameters and to study the efficiency of photodegradation by solar light of pesticides present in honey.

*Bibliographical
part*

Chapter I.
Honeybee,
melliferous
plants and
beekeeping

1. Honeybee

1.1. Classification

Honeybees have been on earth for just over 50 million years, it is a social insect, it lives in colonies of around 50 000 individuals. The average number of a colony must be around 100 000 bees at its maximum, while, 60 000 is the number necessary for a good wintering to reach 30,000 or 40 000 at the end of winter. The more a colony is populated, the more it will harvest honey, for identical exterior conditions (Mathis, 1941). According to Corbara (1991) honeybees appeared around 100 million years ago.

Nowadays, over one million insect species have been described (Stork, 2018; Robert et al. 2009). Among this huge diversity of insects, honeybees are receiving special attention (Koeniger et al. 2010). Honeybees belong to the kingdom Animalia, the phylum Arthropoda, the class Insecta, and the order Hymenoptera (from the Greek hymen, for membrane, and pteron, for wing) (Gupta et al., 2014). Honeybees are a social flying insects and members of the genus *Apis* of Apidae family (Marchenay, 1984). Eleven identified species are forming the genus *Apis* (Crane, 2009). Michener (2000) has classified the honeybee's species as follow:

1. Small species with single exposed combs; dances on expanded horizontal base of comb: *Apis florea* Fabricius (1787) and *Apis andreniformis* Smith (1858).
2. Large species with single exposed combs; dances on vertical curtains of bees or on comb: *Apis dorsata* Fabricius (1793), *A. laboriosa* Smith (1871), *A. binghami* Cockerell (1906) and *A. breviligula* Maa (1953).
3. Middle-sized species with multiple combs in cavities; dances on vertical surfaces of combs in the dark: *Apis mellifera* Linnaeus (1758), *A. cerana* Fabricius (1793), *A. koschevnikovi* Buttel-Reepen (1906), *A. nigrocincta* Smith (1861) and *A. nuluensis* Tinget, Koeniger and Koeniger (1996).

The common name of honeybee (*Apis mellifera*) is the European honeybee or Western honeybee. Carl Linnaeus has given the scientific name of *Apis mellifera* Linnaeus (1758) to honeybee, which means the honey-carrying bee. Later, scientists have proposed the new name *Apis mellifica* (honey-making bee) that seems more accurate; however, the first name is still more utilized (Mattingly, 2012).

1.2. Algerian honeybee' subspecies

Two subspecies within the total of 44 species of honeybees (Engel, 1999) are originally distributed in Algeria. *Apis mellifica intermissa* (Tellian honeybee) was the first geographic subspecies described by Buttel-Repen (1906) (Hepburn, 1998; Gupta et al., 2014). It is a native subspecies of Algeria and its distribution area covers North Africa (Algeria, Tunisia and Morocco), between the Atlas and the Mediterranean and Atlantic coasts (Peng et al. 2016; Shaibi et al. 2009). The second subspecies (i.e. *Apis mellifica saharensis*) has been described successively by Baldensperger (1924) (Haccour, 1961; Cornuet et al., 1988; Gupta et al., 2014). *Apis mellifera sahariensis* is native to Sahara Desert oasis habitats and it is adapted to Saharan flora (Conte and Navajas 2008). *Apis mellifera sahariensis* is originally described in the oases of Western Algeria and Southern Morocco (Shaibi et al. 2009)



Figure 1. *A. mellifera sahariensis* (left picture) and *A. mellifica intermissa* (right picture) entering hives.

2. Melliferous plant

2.1. Introduction

In gymnosperms, the ovum is naked on the spreading carpel (gymno = naked) and the pollen reaches it directly. However, angiosperms have an additional protection, the carpels completely surround the ovum, which is in a closed cavity, the ovary (angio = closed). When the ovary is ripe, it turns into fruit (Roland and Roland, 2003). This includes nearly 253300 species distributed in around 485 families. The angiosperms are divided into two classes, Dicotyledons (196990 species) and Monocotyledons (56310 species), depending on the number of cotyledons per seed (Singh, 2010). Most angiosperm flowers have both stamens and carpels, they are called bisexual or perfect (Simpson, 2006). In this case, the pollen grains released by an anther can be

easily transported and fallen on a stigma of the same flower thus ensuring direct pollination, which leads to self-fertilization. However, this process is often prevented by physical or chemical barriers that separate pollen from the stigma of the same flower (Judd et al., 1999).

The unisexual flowers (imperfect) which carry only one category of reproductive organ (Androecia) with (n) stamen or gynoecium (Pistil) carpels (Singh, 2010). The pollen grains released by a flower can not only deposit on the stigmas of a ripe flower and needs an indirect pollination. Moreover, unisexual flowers can be carried by the same foot, in the case of monoecious species, or by different feet in the rarer species called dioecious which then require cross-pollination (Judd et al. 1999; Singh, 2010). In fact, nothing prevents cross-pollination in almost all types of flowers, which also ensures higher seed quality by introducing high genetic variability. This pollination constitutes a positive selection value and depends above all on the different pollination modes (Miskovsky and Pezold, 1992).

2.2. Definition

Melliferous plant is an entomophilous plant whose flowers are specially visited by honeybees, who come to seek and harvest the raw materials necessary for the survival of the hive and the reproduction of the species, nectar and pollen are two necessary foods, that the honeybee returns to the hive for the production of honey (Marchenay, 1984).



Figure 2. Honeybee foraging a flower of *Taraxacum officinalis*.

2.3. Nectar

Nectar is a sweet and fragrant substance, secreted by nectar-bearing glands, which is found on many plants. In most cases, the nectaries are most often located at the base of the pistil. This is

where bees come by means of their trunks and their tongue draw nectar. Pollen is the main source of protein, minerals, fat and many other substances for honeybees, while nectar provides carbohydrates (Herbert and Shimanuki, 1978). The secretion of nectar is prior to the appearance of flowering plants. In ferns, nectar secretions are already observed, which is why bees occasionally visit female ferns. A nectar called "extrafloral" can also be secreted in flowering plants, away from the flower, on the stems and leaves. These are excess products of photosynthesis oozing from the riddled phloem tubes (Fluri et al., 2001).

Whatever the position of the nectar on the plant, there are two groups, one that produces nectar of phloem sap, and one that gives the end of xylem and phloem sap. The nectar of the latter group is most often rejected by bees because the xylem sap, which is dominant, contains a very low percentage of sugar. Flower nectar is the most important substance used by honeybees to make honey. Therefore, honeybees do not usually harvest the one that contains less than 14% sugar, unless they only have this source at a given time and it is abundant. In general, it should be noted that the chemical composition of the nectar and the amount secreted during the day varies from one plant to another, because the secretion of nectar depends on endogenous factors, namely, the physiological state of the plant, its age, its state of health and its genetic characteristics, and exogenous factors, notably climatic and edaphic conditions (Biri, 1986).

2.4. Pollen

The pollen grain is the male gametophyte of spermatophytes (Douzet, 2007). Pollen is a living sex cell surrounded by two protective layers, the intin and the exin. The cell contains the cytoplasm and two nucleoli which are not visible with the method used for identification. When a pollen grain is deposited on the terminal part of the pistil, it germinates by forming a long pollen tube in which two male gametes are formed. When the end of the pollen tube encounters the ovum, the male gametes penetrate inside, to reach the embryonic sac. The embryo then develops, then the seeds (Lézine, 2011). The pollen grains are simple with a single cell (monad), the most frequent case, composed in tetrad (4 seeds) case of Ericaceae (heather, rhododendron, etc.) or composed in polyades (6 to 8 or 12 adjacent grains), case of Mimosaceae (Lézine, 2011).

The two protective layers of the pollen grain are:

- The intine, which is a thin pectocellulosic membrane similar to the primary wall of plant cells surrounding the pollen protoplast (Shivanna, 2019; Shukla et al., 1998).
- The exine, which is the outer layer of the pollen grain. The exine is made of particularly resistant material, which is found in fossil after millions of years, it is sporopollenin, (Shivanna, 2019; Shukla et al., 1998). According to Faegri (1956) in Jones and Rowe (1999), exine is divided into two layers: endexine and ectexine, the latter consists of three strata: the tectum (sometimes incomplete) columellae (arranged radially more or less separated) and the ground (a light and uniform base).

According to Hideux (1979), exine comes in three forms: Exine complete (complete tectum), exine incomplete (perforated tectum or partial tectum), exine absent (tectum absent). On the other hand, the ornamentation of exine is an essential criterion for the identification of pollen grains from different plant species. It frequently presents geometrical figures or lines which generally allow a good identification, let us quote some types (Reille, 1990; Simpson, 2006; Traverse, 2007):

- Psilate exine (buckthorn), having a smooth sculpturing.
- Foveolate exine (linden), having a pitted surface.
- Striate exine (fruit plum genus), having a fingerprint style sculpture.
- Punctate exine (bellflower), having many small black dots.
- Baculate exine (mistletoe), having a rod-shaped element.
- Reticulate exine (lily, rock rose, rapeseed), in a form of networked or net.

We can see on the pollen surface areas presenting a thinning or even an absence of certain layers of exine, these corresponding to the possible exit point of the pollen tube, these are the apertures. Depending on their shape, a distinction is made between porus (porate pollen) of rounded shape (ulcus if is not equatorial), colpus (colpated pollen) of elongated shape (sulcus if is not equatorial) or a combination between porus and colpus (colporate pollen). Monocolpate (one colpus), dicolpate (two colpi) and triporate (three pori) (Halbritter et al., , 2018). On the other hand, the larch is inaperturate (no pores or furrows) but the borage has 6 furrows (stephanocolpate grain). Some grains are more specific such as *Lychnis* which have pores on the entire surface (peripore grain) or alternating pores and furrows like loosestrife (heterocolporate). The sweet clover which

has three pores (triporate). Apertures appear on the pollen grain in several positions: polar, meridian, equatorial, depending on the plant species (Cerceanu-Larival and Hideux, 1983).

2.5. Pollination

Pollination is the transport of pollen from the male part to the female part (Grassino, 1993). This transport is carried out due to physical factors (gravity, water, wind) or to biological agents (insects, birds or mammals) (Bacher, 2006). The pollen grains germinate on the stigma, then they form a pollen tube that grows through the tissues of the style to the eggs, which are wrapped in the ovaries. It is probably chemicals produced by the egg that guide these pollen tubes. Through these channels, male germ cells migrate to the oospheres. The fusion of male and female sex cells is called fertilization (Fluri et al., 2001).

2.5.1. The different modes of pollination

Pollination has two types i.e. self and cross-pollinations (Lloyd and Schoen, 1992; Yacine and Bouras, 1996). Self-pollination happens in the same plants and cross-pollination occurs between different plants (Mangena and Mokwala, 2018). There are three modes of cross-pollination (Judd et al. 1999):

- By the wind, they are called anemophilous plants.
- By water, they are called hydrophilic plants.
- By animals, they are called zoophilic plants, for instance, entomophilous plants: fertilized by insects, ornithophilous plants: fertilized by birds, cheiropterophilous plants: fertilized by bats and malacophilous plants: fertilized by gastropods.

2.5.2. Pollination by insects

The entomophilous plants depend on pollinating insects which assure the transport of the pollen from the anther of one flower to the stigma of another, on the same or different plant (Bacher, 2006). When an insect visits a flower, the pollen attaches to the bristles of its hairy body and by entering other flowers, the insect involuntarily leaves a few grains of pollen on the stigmas. Flowers attract their pollinators by color and smell (Judd et al. 1999). Insects do not participate equally in pollination. On the other hand, the list of flowering plants pollinated by honeybees thus includes around 170,000 species (Tautz, 2009). Most flowering plants are partially or totally pollinated by honeybees.

Indeed, bees constitute a key element of the ecosystem through its role as pollinator (Cardinaux, 1995).

Flowering plants represent 70% of the plant kingdom, or around 240,000 species worldwide (Bacher, 2006). About 1,000 species of plants can only reproduce due to bees, because they have no other means of carrying out pollination, no other insect, no atmospheric agent being able to ensure it (Ravazzi, 2003). Pollination of wild plants, although more discreet, is just as important, knowing that 80% of flowering plants depend on insects as the carriers of pollen (Marchenay, 1984). On the other hand, reduced forage diversity and abundance, land-use change, pathogens, disease, pesticide exposure, and socio-economic factors are dangerous for the pollinators' survival (Cornman et al., 2015).

2.5.3. Characteristics of the entomophilous flower

2.5.3.1. Factors that ensures the approach and the visit of insects

- Floral envelope consisting of calyx and corolla, which are attractive with clearly visible distinctive signs (color, shape, size, distinctive signs for insects).
- Odor.
- Nectar (Fluri et al., 2001).

2.5.3.2. The floral characteristics which favor the transfer of pollen by insects

- Relatively low pollen production (some 1,000 or 10,000 grains)
- Sticky pollen
- Pollen with high nutritional value (up to 30% protein, 10% fat, 7% starch, vitamins and minerals)
- Pollen with rough surface (Fluri et al., 2001).

In addition, certain anatomical particularities ensure in some species the maximum efficiency in visiting insects for pollination (Fluri et al., 2001).

2.5.3.3. The arrangement of nectaries in the flowers

Nectaries are clusters of small glandular cells, surrounded by thin cellulosic walls. Nectar Can be secreted in flowering plants, away from the flower, on stems and leaves (Fluri et al., 2001). Nectar is the greatest reward for pollinators when they visit flowers. In all cases, the nectaries are

located deeply, forcing the foraging insect to penetrate through, which always favors the harvesting or deposition of pollen (Holm, 1979).

2.5.4. Variation and condition of the pollen supply.

The honey capacity varies greatly with the family, genus and species of the plant. Even in a given species, it can vary greatly from one variety to another (Philippe, 1988; Winston, 1993). Pollen is generally available in the morning, but some plants have anthers that open at any time of the day and even at night (Louveaux, 1958). The production of pollen by the plant is genetically determined, but the influence of the environment is important, temperature and humidity are essential factors. Maturity is advanced by the heat while the dehiscence (opening) of the anthers is favored by a certain dryness of the air (Guerriat, 2000).

Table 1. Rate of pollen supply of some plants (Guerriat, 2000).

Species	Rate of pollen supply
Anemone and buttercup	Especially from 10 to 12 a.m.
Chestnut	From 7 to 7p.m. with a peak from 9 to 11 a.m. and 3 to 5 p.m.
Clematis	Especially from 8 to 10 a.m.
Poppy	Especially before 10 a.m.
St. John's Wort	From 6 to 12 p.m., especially early in the morning
Dandelion	50% of the balls harvested between 7 and 9 a.m.
Pear tree	Especially in the afternoon
Groundsel	8 to 5 p.m., especially between 10 and 12 p.m
Clover	Maximum between 10 a.m. and 2 p.m.

2.6. The relationships between the bee and the flower

The relationship between the honeybee and the flower is based on reciprocal exchange, food source (nectar and/or pollen) advantage for the bee and reproduction advantage for the plant. Superior angiosperms have been established and diversified the relationship with their pollinating organisms (Pesson and Louveaux, 1984).

2.6.1. The relationship of the honeybee with the flower

Feeding the honeybee colony is a doubly complex process. We find at the level of the individual the usual functions of digestion, assimilation, excretion. At the colony level, different functions appear. Food collection is done by a category of specialized individuals, foragers. In one year, the foragers of a normal-strength colony harvest a hundred kgs of nectar and 30 to 50 kg of pollen (Louveaux, 1968).

The staple foods of the colony are nectar and pollen. The nectar by its composition mainly provides sugars and water. Nectar and pollen are stored; the nectar stored in the shelves becomes honey by losing its excess water and enriched with enzymes from salivary secretions. Stored pollen in the rays also undergoes a lactic type fermentation. pollen is deeply modified and enriched (Pain and Maugenet, 1966 in Guettar, 2006).

2.6.2. The relationship of flowering plants with the honeybee

Many flowering plants are dependent on insects for their pollination. This phenomenon has enabled the creation of a biological association. All good melliferous plants are characterized by very well developed floral and extra nectaries, which can be concentrated and secrete sugars, and their flowers generally are adapted to attract bees. Some plants produce little or no nectar but are more attractive to the bee because of their pollen production. The practice of beekeeping necessarily deserves an elementary knowledge of melliferous plants, their physiology (nature and quality of their nectar and pollen production), ecology, distribution, influence of environmental factors (Louveaux, 1980). About fifty species of cultivated plants represent almost half of major endogamous food plants, and therefore they need insects for their pollination and fruiting, in particular honeybees (Philippe, 1991). The genus *Apis* is the most effective pollinating insect, not only by the rigorous adaptation of its morphology to the collection of nectar and pollen, but also by the large number of individuals that constitute a colony (Tautz, 2009).

3. Beekeeping

3.1 History

The domestication of the bee dates back around 6000 years. The use of beehive products is reported in the time of the Pharaohs, 3600 years BC in Egypt. Earthen hives, made 3400 years BC, were discovered in Crete, at Phaistos and Knossos (Vaillant, 1991).

Representations of beehives dating from 2500 years BC have been found in the western Mediterranean. In the Ancient Egyptian Empire, an apiary formed of stacked pottery and scenes depicting the extraction and conservation of honey are testimony to flourishing beekeeping 2400 years BC (Corbara, 1991).

3.2. Beekeeping advantages

3.2.1. Agronomic advantages

By foraging in search of nectar and pollen, the honeybee actively participates in the pollination of wild flora (hawthorn (*Crataegus oxyacantha*), dog rose (*Rosa canina*), mountain ash (*Sorbus domestica*) ...), but also cultivated plants, promoting their reproduction and improving harvests (Williams, 2000). The usefulness of honeybees has been gradually recognized by many countries such as the USA and the USSR, and foresee the need to increase the number of hives per hectare in order to increase the unit production of their crops and ensure the conservation of nature (Guerriat, 2000).

Without pollination, no fertilization so no fruit or vegetables. Eighty percent of plant species need honeybees to be fertilized (Vannier, 2005). As a general rule, the proximity of beehives increases productivity more than a one-third (Cherbuliez and Domerego, 2003). In California, honeybees are essential to pollinate the 285,000 hectares of almond trees (Lindsey, 2008). On the hand, researchers attach great importance to the pollination action of honeybee and they consider it as a vast field of research and new discoveries (Biri, 1997).

3.2.2. Economic advantages

Apis mellifera is as essential to the economy as it is to the survival of the human species (Vannier, 2005). Beekeeping can be an important speculation in the case of cooperation or professional producers by the production of a whole range of products (honey, jelly, wax, venom, swarms, queens, etc.). The economic contribution of bees to world agriculture is estimated at 117 billion US \$ (about 12 trillion Algerian Dinar) (Costanza et al., 1997). The beekeeper can be used

as a factor to enhance the value of different crops due to the pollination which allows an increase in quality and quantity (Biri, 1997).

3.2.3. Therapeutic advantages

Since humans have used plants to fight diseases or heal wounds, their observations of the life of honeybees led him to use their products and especially honey against various diseases. Honey was already appreciated by the Romans who used it as a food as well as a medical and cosmetic ingredient (Biri, 2002). Due to its medicinal action, honey plays a very important role and it is used against sore throats, diarrhea, etc. In addition, the royal jelly has constituents, endowed with erythropoietic, granulopoietic and thrombopoietic properties (Cherbuliez and Domerego, 2003). Moreover, pollen and bee bread have an effect on the aging process, generally causing memory loss. Pollen is the richest food compound in Selenium (antioxidant) (Cherbuliez and Domerego, 2003).

3.3. The beehive

Humans have started using the hollow tree trunks as beehives, which are closely modeled the natural condition of bees' life in the wild. Later, the needs of men are increasing, they have been led to develop more practical beehives that is easier to achieve and using materials that are easy to find and of little value. From the antiquity to the present day and depending on the place of the globe, the shape of the hives has varied considerably (hollowed trunks, terracotta, straw, wood, etc.) (Adam, 1980).

3.4. The Apiary

The apiary is made up of all the hives gathered by a beekeeper in a specific location. The types of apiary are outdoor apiary, covered apiary, chalet apiary and transhumant apiaries (Guerriat, 2000).

According to Warré (2015), recommendations for better productivity of apiary are:

- The shade is necessary for the hives because the sun melts the wax and the honey, destroys combs and drowns bees. Moreover, it stops honeybees from going out and forces them to ventilate the hive.
- It is recommended to orient the hives East if not available they can be faced West or North but never South, which helps to awaken the foragers earlier by the sun.

- The hives should be at least two meters away from a wall of two meters high.
- The number of hives should fit the size of land.
- It is important to leave distance of four to six meters between the hives.
- The number of hives in an apiary should be in proportion to the nectar supply in the locality.
A number 50 hives within a radius of three kilometers is recommended.
- The beekeeper can plant melliferous plants near his hives to support nectar supply.
- Never allowed near the apiary the following plants: tobacco, belladonna, henbane, hemlock, aquilegia, hellebore, rose-laurel, foxglove, thorn-apple, monkshood, varnish- tree, autumn crocus. These plants are not all harmful to bees, but their alkaloids pass into honey which then becomes dangerous.

3.5. Beekeeping value of the plant cover

The study of plant cover allows the beekeeper to evolve the beekeeping value of the environment of his apiary. The composition of the plant cover in the foraging area around the apiary influences the harvesting and development potential of the colonies. Ideally, the foraging area should provide the colonies with enough resources throughout the year, for both pollen and nectar. When the colonies are strongest, the nectariferous secretion in the foraging zone must allow the colonies to realize significant reserves of honey, which requires a vegetal cover where the nectariferous plants are abundant (Guerriat, 2000).

The first phase of the study consists in delimiting the foraging area of the apiary. This is defined by the surface explored by the honeybees around their apiary. The second phase consists of traveling through the region and writing down on a map the main types of vegetation encountered (Guerriat, 2000).

3.6. Variation in honey production

On a beautiful day, a third of a colony (which can represent 15,000 - 20,000 bees) can leave the hive to forage and produce up to 6 kg of honey. For every kilo of honey, it takes almost 50,000 flights and more than a million visited flowers. This makes it clear that during their life as a forager, each worker specializes in a floral species until the depletion of the resource or the identification of more interesting resources. These figures illustrate the quantitative and qualitative importance of pollination by the honeybee (Cherbuliez and Domerego, 2003).

3.6.1. Intensity of foraging

Rabiet (1989) notes that when a plant is foraged, its exploitation continues if it can provide one of the desired products. He explains this law by the fact that, when honeybees find a source of nectar, they exploit it until complete exhaustion. On the other hand, Marchenay (1988) reported that the production of nectar is constantly renewed due to its pollination.

3.6.2. Soil

Hommel (1947) shows that the texture of the soil has a considerable influence on the intensity of the nectariferous secretion. For instance, sainfoin is more melliferous in calcareous soils than sandy soils. White mustard becomes more melliferous on sandy limestone soils and secretes less nectar on clay soils.

3.6.3. The light

The sun light stimulates a lot of flowers to open, which stimulate honeybees to visit them. Bees preferably visit well-lit flowers and abandon them to pass over others following the movement of the sun (Hommel, 1947).

3.6.4. The climate

The climate is a very important element which conditions the nectariferous secretion (Jean-Prost, 1987). Several non-melliferous plants in certain regions can become so in others, depending on the presence or absence of favorable conditions. In general, it has been shown that the succession of several days of good weather and rainy weather at the time of flowering promotes the production of nectar (Signorini, 1979; Louveaux, 1980).

3.6.5. The temperature

Temperature is a limiting factor for nectar-producing secretion. In apricot and acacia, the nectar-bearing activity takes place only at temperatures of 15 °C and 18-20 °C, respectively (Louveaux, 1980).

3.6.6. The air humidity

The amount of nectar can increase or decrease according to the hygrometric state of the air. Certain plant species such as willowherb and orpins are nectariferous in dry weather (Marchenay,

1988).

3.6.7. The latitude

The volume of nectar emitted increases with latitude at least for spontaneous plants (Louveaux, 1980). Therefore, honey production by honeybees will be affected by the latitude location.

3.7. The floral calendar

3.7.1. Definition

A floral calendar is the timetable that indicates to the beekeeper the approximate date and duration of the flowering periods of the plants in his area. It requires full observation of seasonal changes in the vegetation patterns of the area's agro-ecosystems, and the foraging behavior of honeybees and the way in which honeybee colonies interact with their floral environment. Preparing a precise and detailed calendar will often require several years of repeated recording and refining of the information obtained (Guerriat, 2000).

3.7.2. The stages of making a floral calendar

According to Guerriat (2000), the steps taken to make a floral calendar are:

- Make a general survey of the sector, write a list of flowering plants found and estimate with attention the density of floral populations.
- Place several honeybee colonies in the area with regular inspection of the supply status of the hives.
- Determine if plants are targeted for nectar or pollen.
- Study the frequency with which the bee visits each flower species in relation to changes in the feeding level of colonies.
- Carefully record all flowering changes, noting that at the end of flowering, colony plants begin to lose weight.

Chapter II.

Beekeeping

products

1. Honey

Honey constitutes the energy reserve of the hive. We can evaluate the honey consumption of a honeybee at 120-170 mg per day, knowing that it can be up to 200,000 births per year in a well-nourished colony, this corresponds to an annual consumption of 30-40 kg of honey (Cherbuliez and Domerego, 2003). Knowledge of honey and its origin has retained for long time a mystical value. Honey has always been a sacred product due to these precious virtues (Gonnet, 1982).

According to Codex Standard for Honey (2001) “Honey is the natural sweet substance produced by honey bees from the nectar of plants or from secretions of living parts of plants or excretions of plant sucking insects on the living parts of plants, which the bees collect, transform by combining with specific substances of their own, deposit, dehydrate, store and leave in the honey comb to ripen and mature.”

1.1. Chemical composition of honey

1.1.1. Sugars

Sugar content is about 80–83% in honey, composed principally by fructose and glucose and smaller amounts of near 30 of other sugars (Alvarez-Suarez, 2017). In honey, monosaccharides are dominant with about 75%, disaccharides represent 10–15% and small amounts of other sugars have been found (Da Silva et al. 2016). Fructose plus glucose content of over 60% for floral honey and 45% for honeydew honey and blends of honeydew honey with floral honey is required by both Codex and Directive (Thrasyvoulou et al., 2018). Sugars in honey determine properties such as energy value, viscosity, hygroscopicity and granulation (Da Silva et al. 2016). According to Alvarez-Suarez (2017), sugars that have been identified in honey are:

- Monosaccharides: Glucose, fructose and galactose (occasionally cited in trace levels).
- Disaccharides: Majority level are isomaltose, kojibiose, maltose, sucrose and turanose; minority level are cellobiose, gentiobiose, maltulose, nigerose and palatinose; traces level are isomaltulose, laminaribiose, leucrose, melibiose and trehalose.
- Trisaccharides: Majority level are erlose, theanderose, panose and maltotriose; minority level are isomaltotriose, isopanose, melezitose and raffinose; traces level are centose, 1-kestose, laminaritriose, planteose and α -3'-glucosyl-isomaltose.

- Higher oligosaccharides: Traces level are isomaltotetraose, maltotetraose, isomaltopentaose and nystose.

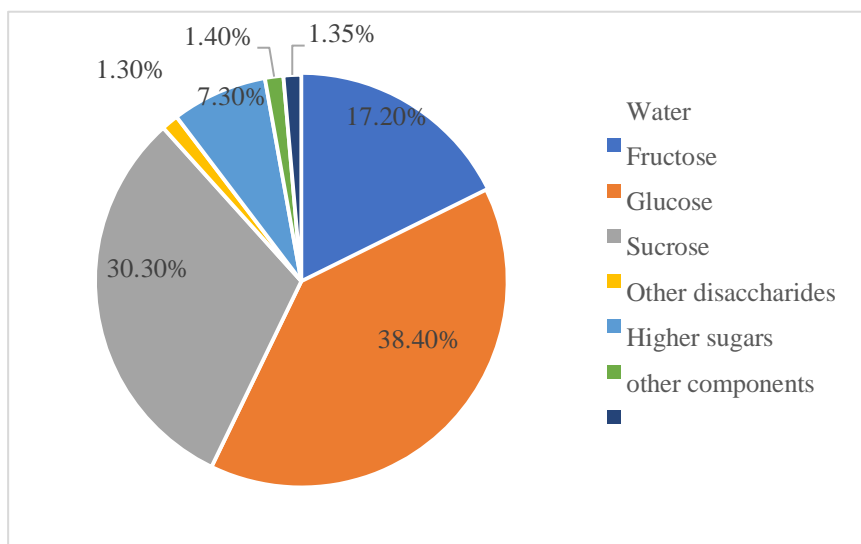


Figure 3. Chemical composition of U.S. honey (Ball, 2007).

1.1.2. Amino acids and proteins

Animal and vegetal sources, including fluids and nectar secretions of the salivary glands and pharynx of honeybees, are the origin of proteins in honey (Da Silva et al., 2016). However, the pollen is the major source of proteins, lipids, vitamins and minerals for the honeybee (Lamontagne-Drolet et al., 2019). The amount of protein content in honey is affected by honeybee species produce it, for instance, proteins represent between 0.2% and 1.6% of *Apis mellifera* and 0.1–3.3% of *Apis cerana* honeys. In addition, different foraged plants give different amount of proteins in honey, eucalyptus honey (0.6%), blackberry or polyfloral honeys (0.7%) and manuka and heather near to 1.5%, which elevate honey viscosity (Alvarez-Suarez, 2017). The proteins present in honey can be used to test the honey authenticity, adulteration and quality (Bocian et al., 2019). The carbohydrate metabolism enzymes and the royal jelly characteristic proteins are major groups of proteins to be found in honey. The royal jelly characteristic proteins include major royal jelly proteins 1–5 (mostly MRJP1), royalisin (known as defensin-1) and apisimin. The

carbohydrate metabolism enzymes include glucose oxidase, invertase and diastase (Lewkowski et al., 2019).

Table 2. Main honey enzymes and their activities (Alvarez-Suarez, 2017).

Enzymes	Activity	
α -Glucosidase (invertase)	Converts sucrose to glucose and fructose (inverts sugar)	7.5–10 g saccharose hydrolyzed by 100 g honey per hour at 40 °C
α - and β -amylase (diastase)	Transform starch to other carbohydrates (dextrins, oligo-, di- and monosaccharides)	16–24 g starch degraded by 100 g honey per hour at 40 °C
Glucose oxidase	Converts glucose to gluconolactone, which in turn yields gluconic acid and hydrogen peroxide	80.8–210 μ g H ₂ O ₂ formed per g honey/h
Catalase	Converts hydrogen peroxide to water and oxygen	0–86.8 catalytic activity/g honey
Acid phosphatase	Removes phosphate from organic phosphates	5.07–13.4 mg P/100 g honey released in 24 h
Proteases	Hydrolyze proteins and polypeptides to yield peptides of lower molecular weight	
Esterases	Break down ester bonds	

Table 3. Average contents of free amino acids in honey (Alvarez-Suarez, 2017).

Amino acid	mg/100 g honey (dry weight basis)	Amino acid	mg/100 g honey (dry weight basis)
Asp (aspartic acid)	3.44	Tyr (tyrosine)	2.58
Asn + Gln (asparagine +glutamine)	11.64	Phe (phenylalanine)	14.75
Glu (glutamic acid)	2.94	β -Ala (β -alanine)	1.06
Pro (proline)	59.65	γ -Abu (γ -aminobutyricacid)	2.15
Gly (glycine)	0.68	Lys (lysine)	0.99
α -Ala (α -alanine)	2.07	Orn (ornithine)	0.26
Cys (cysteine)	0.47	His (histidine)	3.84
Val (valine)	2.00	Trp (tryptophan)	3.84
Met (methionine)	0.33	Arg (arginine)	1.72
Ile (isoleucine)	1.12	Others	24.53
Leu (leucine)	1.03	Total	118.77

Most of the amino acid content originates from the honeybees and not from the nectar or the pollen (Ball, 2007). Twenty-six amino acids have been reported in honey. Amino acids represent between 0.3 and 1 % (w/w) of the total honey weight. Proline is the dominant amino acid in honey and its amount can be used to report the total amino acid content in honey, since the other amino acids are present in trace quantity (Chua et al., 2013).

1.1.3. Organic acids

Organic acids represent about 0.5% of fresh honey, and they are originally contained in nectar or derived from sugars when transforming nectar to honey by enzymes of honeybees (Alvarez-Suarez, 2017). Organic acids have a big impact on organoleptic properties (color and flavor) and physical and chemical properties (pH, acidity, and electrical conductivity) of honey (Mato et al., 2003). In addition, they contribute in antibacterial and antioxidant activities of honey (Mato et al., 2006). 32 organic acids have been reported in honey from different regions (Alvarez-Suarez, 2017) : aspartic acid, butyric, citric, acetic, formic, fumaric, galacturonic, formic, gluconic, glutamic, glutaric, butyric, glyoxylic, 2-hydroxybutyric, a-hydroxyglutaric, isocitric, aketoglutaric, lactic, malic, malonic, methylmalonic, 2-oxopentanoic, propionic, pyruvic, quinic, shikimic, succinic, tartaric, oxalic and others (Da Silva et al., 2016). Moreover, organic acids can be used to discriminate between honeys according to their botanical and/or geographical origins, as fermentation indicators or for the treatment of Varroa infestation (Mato et al., 2006).

1.1.4. Phenolic compounds

Phenolic compounds represent important groups of secondary metabolites biosynthesized by plants as a defensive tool against biotic and abiotic stress and oxidative damage (Alvarez-Suarez, 2017). The nectar is the main source of over 200 polyphenol compounds have been identified in different types of honey (Jibril et al., 2019). Using unspecific methods like Folin-Ciocalteu, the total phenolic content (TPC) in honey has been reported to be ranged between 20 and 193 mg gallic acid equivalents (GAE)/100 g of honey (Alvarez-Suarez, 2017). The TPC in honey varies depending on the plant source, and darker honey tends to have higher TPC than lighter honey (Molaveisi et al., 2019). Phenolic compounds have been known as the main responsible for the antioxidant activity of honey due to the ability to scavenge or reduce the formation of free radicals (Da Silva et al., 2016; Cianciosi et al., 2018). In addition, the major tow

phenolic compounds groups found in honey are flavonoids (apigenin, chrysin, galangin, hesperetin, kaempferol, luteolin, myricetin, pinobanksin, pinocembrin, quercetin, and tricetin) and phenolic acids (caffeic, chlorogenic, coumaric, ellagic, ferulic, gallic, homogentisic, phenyllactic, protocatechuic, syringic and vanillic acids) (Campone et al., 2014). Moreover, Da Silva et al. (2016) reported that the main functional components of honey are flavonoids. They can significantly contribute to the total antioxidant activity of honey, bringing beneficial effects for human health. On the other hand, the antioxidant activity of flavonoids mostly depends on the number and position of hydroxyl groups and other substituents and the glycosylation of flavonoid molecules. Therefore, the analysis of phenolic compounds is important to evaluate the quality of honey, which is judged by its botanical origin and nutraceutical value (Campone et al., 2014). Finally, phenolic compounds are subjected to degradation by the environmental conditions (Da Silva et al., 2016).

Table 4. Phenolic compounds found in honey (Abubakar et al., 2012).

Category	Compound
Flavonoles	Quercetin, kaempferol, Galangin, Fisetin, Myricetin
Flavanones	Pinocembrin, Naringin, Naringenin, Hesperidin Pinobanksin
Flavones	Apigenin, Acacetin, Chrysin, Luteolin Genkwanin, wogonin, tricetin
Phenolic acids	Caffeic acid, chlorogenic acid, cinnamic acid, p-coumaric acid, vanillic acid, ferulic acid, p-hydroxybenzoic acid, gallic acid, syringic acid, rosmarinic acid and derivatives
Coumarins	Coumarin
Tannins	Ellagic acid

1.1.5. Volatile compounds

The aroma profile is a key feature to characterize the organoleptic quality and authenticity of a food product. Volatile compounds (VCs) have the main responsibility in the determination of the aroma and flavor of foodstuffs. Honey contains various volatile and semi-volatile compounds that depict its flavor and fragrance qualities (Karabagias et al., 2020; Manyi-Loh et al., 2011). In addition, volatile and semi-volatile compounds can participate in the antimicrobial and some therapeutic properties of honey (Moniruzzaman et al., 2014). The volatile composition of honey depends mostly on the nectar composition and floral source (Viuda-Martos et al., 2010) and varies also according to climate, soil, age of honey, mode of storage, honey processing and bee species (Makowicz et al., 2018).

Aromatic compounds are found in very small quantities in honey (Bianchin et al., 2014). In honey, over 600 volatile compounds have been reported by authors (Karabagias et al., 2020; Manyi-Loh et al., 2011). Terpenes and their derivatives, norisoprenoids, and benzene derivatives are the major well-known categories of honey volatiles (Pattamayutanon et al., 2017). Some volatile classes include esters, ethers, alcohols, carboxylic acids, aldehydes, carotenoid derivatives hydrocarbons, ketones, terpenes, nor isoprenoids, furan and pyran derivatives and phenolic volatiles (Karabagias et al., 2020). Volatile compounds in honey can be originated from the plant source, from honeybees by producing new ones or converting plant constituents to different volatile compounds or from postharvest processing by the intervention of micro-organisms present in honey after. Some authors have found modification in the volatile profile during the storage of honey. For instance, the disappearance of 17-pentatriacontene in cashew honey was reported by Moreira et al. (2010) and Kaskoniene et al., (2008) have found that the amount of octane increased with storage time (Da Silva et al., 2016). Volatile compounds have been utilized to differentiate honeys from different geographical origin (Patrignani et al., 2018).

1.1.6. Vitamins

Vitamins are among different biological bioactive compounds present in honey (Meo et al., 2017). Vitamins and minerals in honey are found in small quantities and they have a marginal contribution in the recommended daily intake (RDI) of the different trace substances (Bogdanov et al., 2008). In honey, most of vitamins are originated from pollen and mainly are water-soluble with the predominance of Ascorbic acid (C), which is practically present in different kinds of honey with a concentration of about 2 mg/100 g. Other vitamins are thiamine (B1), riboflavin (B2), nicotinic acid (B3), pantothenic acid (B5), biotin (B8 or H) and folic acid (B9) (Alvarez-Suarez, 2017) and phylochinon (K) (Alvarez-Suarez et al., 2010a).

Low pH of honey helps to preserve available vitamins. On the other hand, two causes can reduce the content of vitamins in honey. The first one is the filtration of honey usually for commercial purposes, which due to pollen removal. The second one is the oxidation of ascorbic acid by the hydrogen peroxide produced by glucose oxidase (Da Silva et al., 2016).

1.1.7. Minerals

The mineral content of honey varies between 0.04 and 1.03% and dark honeys have more minerals than light honeys (Bogdanov et al., 2007). Up to 54 minerals have been reported in honey, while potassium is the most abundant one representing one-half to three-quarters of total mineral content (Da Silva et al., 2016; Alvarez-Suarez, 2017), including major (Ca, Mg, Na, K, Cl, P, S) and trace minerals (Zn, Al, Mn, Pb, Cd, Cu, Tl, Co, Rb, Ni, Ba, Bi, Be, Pt, V, Pd, U, Fe, Te, Mo, Hf, Sb, Sn, La, Sm, I, Tb, Dy, Th, Sd, Nd, Pr, Lu, Yb, Gd, Er, Ho, Ce, Cr, B, As, Br, Cd, Se, Hg and Sr (Lanjwani and Channa, 2019). The natural absorption of minerals by plants from the soil and the environment is the major source of the mineral content in honey. On the other hand, the artificial absorption can also contribute in the mineral content in honey originating from artificial sources such as sugar or syrup fed on by the honeybees (Moniruzzaman et al., 2014). The main source of minerals in honey is the botanical origin, however, other factors can contribute such as environmental pollution, beekeeping practices and honey processing (Lanjwani and Channa, 2019). The amount and variety of minerals in honey varies according to their availability in the plants and the soil and soil and environmental contaminations (Kędzińska-Matysek et al., 2018).

Minerals essential to the proper functioning of the human body are present in honey, among them components of compounds that influence metabolism, participate in water electrolyte balance, and have a regulatory effect (Kędzińska-Matysek et al., 2018). However, high levels of trace minerals lead to dangerous effect in the humans due to the incapability of completely metabolizing minerals by human body viz the minerals will be gathered in human tissues without completely destroyed or inactivated (Lanjwani and Channa, 2019). The maximum residue levels of these potentially toxic elements in honey have not been established. On the other hand, a proposal to set up acceptable levels of 15 lg kg/1 for arsenic, 25 lg kg/1 for lead, 5 lg kg/1 for mercury and 7 lg kg/1 for cadmium by World Health Organization (WHO) and the Food and Agriculture Organization (FAO) have been established (Da Silva et al., 2016).

1.1.8. Pesticides

More than 150 pesticides of different types have been found in colony samples and the varroacides are most reported, especially, the acaricides bromopropylate, coumaphos, and fluvalinate. Residue levels increase from honey to pollen to beeswax (Al-Waili et al., 2012). Pesticides can be transmitted to honeybees by consumption of pollen and contaminated nectar, contact with contaminated plants and soils, inhalation during flight and recollection, ingestion of

polluted surface water and direct overspray or flying through spray drift (López et al., 2014). The pesticides used by farmers cause mortality of bees and contamination of beekeeping products, especially in spring and summer (Bargańska et al., 2016). Pesticides are toxic and they present a potential carcinogenic danger can lead to chromosomal abrasions (Darko et al., 2017). Depending on the actual toxicity of the chemical as well as the length and level of exposure, pesticides can cause mild skin irritation, birth defects, tumors, genetic changes, blood and nerve disorders, endocrine disruption, and even coma or death (Al-Waili et al., 2012; Abdallah et al., 2017). According to the World Health Organization (WHO), there are one million serious pesticide poisonings worldwide each year, resulting in about 220000 deaths a year (Cherin et al. 2012). Many studies have revealed the contamination of honey by pesticides, fortunately, most of quantities reported do not present a risk to human health. Maximum Residue Limits (MRLs) through Regulation (EC) 396/2005 were established by the European Union for many pesticides used in the agricultural and apiculture practices (Leu et al. 2010).

Table 5. Pesticide residues reported most frequently in honey (López et al., 2014).

Pesticides	Concentration µg/kg	Country of origin of honey
Organohalogens	0.1 - 4310	Brazil, Turkey, Spain, Portugal and India.
Organophosphates	2.4 – 243	Brazil, China, France, India, Portugal, Spain and Turkey.
Organonitrogen	0.05 – 116	Brazil, Belgium and France.
Pyrethroids	1 - 92	Brazil, China, India and poland.
Carbamates	1 - 645	China, Portugal and Spain.

1.2. Physico-chemical properties

1.2.1. Moisture

The limit for moisture content of honey is set as less than 20% by Codex and European Directive, however, heather honey (*Calluna vulgaris*) can have up to 23% moisture content (Thrasylvoulou et al., 2018). The nectar, harvesting season and beekeeping practices affect the moisture content of honey, which affects honey fermentation and crystallization (Al-Ghamdi et al., 2019). High water content leads to high potential of fermentation and spoilage of honey (Chen, 2019; Singh and Singh, 2018).

1.2.2. Acidity

European Commission set the maximum limit of 50 meq/kg for free acidity of honey (Bouhlali et al., 2019). Gluconic acid, aromatic acids, aliphatic acids are mainly responsible for honey acidity, however gluconic acid, which is originated from glucose oxidase activity during honey ripening, is the most contributor (Laaroussi et al., 2020). The acidity prevents spoiling of honey by microorganisms, and total acidity is a useful indicator of deterioration caused by storage and testing the purity and authenticity. (Suárez-Luque et al., 2002). Moreover, honey acidity increases during storage and fermentation due to the transformation of sugars and alcohols into acids by yeasts activity (Alvarez-Suarez, 2017).

1.2.3. pH

Honey has an acidic nature with a pH level ranging between 3.20 and 4.50 (Da Silva *et al.*, 2016). Honey influences honey texture, stability and shelf life, for instance, low pH prevents the growth and proliferation of microorganisms in honey (Boussaid et al., 2018). Generally, plant source, soil, inorganic molecules, and the honey ripening process can affect the pH level of honey (Ribeiro *et al.*, 2014).

1.2.4. Electrical conductivity and ash

The electrical conductivity is a measure of quality of honey and determined by mineral content and acidity due to ions, organic acids and proteins (Da Silva *et al.*, 2016). The electrical conductivity is used to distinguish floral honey (< 0.8 mS/Cm) from honeydew honey (> 0.8 mS/Cm) (Thrasyvoulou et al., 2018).

Ash content of honey represents the mineral content of honey and is originated from the soil used by flowers to produce the nectar. The ash content can be used as an indicator of geographical origin and environmental pollution (Da Silva *et al.*, 2016).

1.2.5. Hydroxymethylfurfural (5-HMF)

Hydroxymethylfurfural (HMF) is a furanic compound indicating honey freshness. It is made from dehydration of sugars in acidic conditions (caramelization) throughout heat treatment of food as an intermediate in the Maillard reaction (Pasias *et al.*, 2017). According to Codex 2001,

the maximum permitted limit of HMF in honey is <40 mg/kg (Thrasyvoulou et al., 2018). The chemical characteristics such as pH, free acid content, total acidity, lactone content and mineral content are the main factors affect the HMF formation (Shapla et al., 2018).

1.2.6. Diastase activity

Diastases (α - and β -amylase) are among main enzymes found in honey. They are a group of starch-digesting enzymes. The enzyme α -amylase hydrolyses starch chains at random locations, producing a variety of dextrans, while β -amylase splits the reducing sugar maltose from the end of the starch chain (Sak-Bosnar and Sakac, 2012; Sajid et al., 2019). Diastase in honey is either secreted directly by bee's salivary glands or came from nectar or pollen (Sajid et al., 2019). Diastase activity content are well used as criteria to assess the quality of the product and it is sensitive to the heat (Pasias et al., 2017). Diastase activity is expressed as the diastase number (DN) in Schade units and is defined as follows: one diastase unit corresponds to the enzyme activity of 1 g of honey, which can hydrolyse 0.01 g of starch in 1 h at 40 °C ((b) Sak-Bosnar and Sakac, 2012). According to the Honey Quality and International Regulatory Standards, the diastase activity limits has been set to > 8 and > 3 for honeys with naturally low enzyme content (Huang et al., 2019).

1.2.7. The density

The density of a homogeneous honey is the ratio expressed in decimal number of the density of pure water and it varies with water content and temperature (Gonnet, 1982). According to Gonnet (1982), the density of ripe honeys is 1.39 to 1.44 at 20 °C, however, the density can go up to 1.52, in this case the honey is frozen and with butter-like consistency. In fact, the more water in honey, the less dense it is (Hommel, 1947; Abdulkhaliq and Swaileh, 2017).

1.2.8. The viscosity

Viscosity is the ability of a mixture to flow; the higher it is, the slower the flow. The main factors that determine the viscosity of honey are water content, temperature and chemical composition (Chauvin, 1962; Gómez-Díaz et al., 2009; Trávníček et al., 2012). Therefore, higher temperature and content of water in the honey leads to lower viscosity and higher fluidity (Gómez-Díaz et al., 2009; Trávníček et al., 2012).

1.2.9. Color

Color is a very important characteristic of honey and has an important consideration for the consumers (Quintero-Domínguez et al., 2018), usually, light honeys are more expensive in the market (Szabó et al., 2016). Color of honey varies from clear white color to dark amber according to the food source (Lewoyehu and Amare. 2019). Darker honey has mostly more pronounced flavor than light color honey (Manyi-Loh et al., 2011). Water, saccharides, minerals, polyphenols, carotenoids and especially flavonoids strongly influence the color of honey (Selvaraju et al., 2019; Escuredo et al., 2019). Color intensity has a strong correlation with the antioxidant properties of honey, darker honey generally have higher antioxidant properties (Selvaraju et al., 2019).

1.2.10. The specific heat

The specific heat of honey is 0.54 that of water at 20 ° C when the honey contains 17% water (Louveau, 1985). This means that it takes approximately half the energy (of joule) to heat honey than to heat the same mass of water. Honey transmits the heat very badly so that it can be quickly warmed up at one point and stays cold nearby (Jean-Prost, 1987). According to Jean-Prost (1987), honey is 14 times less conductive than water.

1.3. Biological properties

Various bioactive compounds endow honey with antibacterial, antioxidant and anti-inflammatory properties (Gośliński et al., 2019). Phenolic acids, flavonoids, certain enzymes, ascorbic acid, carotenoid-like substances, organic acids, Maillard reaction products and amino acids and proteins promote the antioxidant potential of honey (Khalil et al., 2010). In addition, honey shows antibacterial activity against several pathogenic bacteria, especially gram-positive bacteria, such as *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas* spp. High sugar content, low water activity, hydrogen peroxide, the presence of strong acids, flavonoids and phenolic acids, methylglyoxal, bee defensin-1 and MRJP1 (Lewkowski et al., 2019) are the main factors responsible for the antibacterial power of honey (Dzuga et al., 2018). Likewise, Ahmed et al. (2018) have mentioned the antioxidant, antibacterial, antifungal, Antiviral, Anti-Inflammatory, antidiabetic, antimutagenic, anticancer, antiproliferative, immunomodulatory, cardiovascular effects of honey.

The minor constituents attribute dietetic and medicinal properties for honey. It is an energetic food due to its carbohydrate content and cures several diseases of the digestive system. It acts as a regulator of intestinal functions and contains substances that are very active against pathogenic germs in the stomach and intestines. In addition, it has been confirmed due to animal experiments that honey activates fattening, increases fertility and delays the onset of cancer (Jean-Prost, 1987).

1.4. Honey of nectar

In general, honey of nectar is separated into two distinct categories: mono floral honeys and poly floral honeys (Clément, 2002).

1.4.1. Unifloral honeys (monofloral)

Unifloral honeys are also called vintage honeys (Clément, 2002). Some foraged plant species allow the harvest of monofloral honeys (lime (*Tilia*), acacia (*Robinia pseudoacacia*), lavender (*Lavendula*), dandelion (*Taraxacum officinale*) Monofloral honeys are characterized by the dominance of one plant species (100-70% to be able to be marketed as monofloral honey), which determines precise characteristics as to their taste and appearance, as well as certain properties (Bacher, 2006). Honeybees foraging is rarely limited to one type of flower (Goût and Jardel, 1998).

The most important unifloral honeys are (Loiriche, 1984):

- Rapeseed honey is produced from the nectar of *Brassica napus* var. *oleifera*.
- Acacia honey, which originates from *Robinia pseudoacacia* L., this honey is clear and fine.
- Rosemary honey, Rosemary produces fine honey with a delicate aroma.
- Lavender honey is produced by the species and subspecies of lavender and their hybrids (lavandins), lavender honey is richer in pollens than lavandin honey, it is more colorful.
- Heather honey, from calluna heather, *Calluna vulgaris* L., form a large stand on siliceous ground, its nectar is the source of honey with a full-bodied flavor, slightly bitter, with a powerful odor and medium reddish colors.

The purity of these honeys naturally depends on the extent of the stand of the considered plant (Philippe, 1988).

1.4.2. Multi-floral honeys or poly-floral honeys

Poly-floral honeys are also called all-flower honeys. These honeys are the most numerous, their composition is of course variable and complex, since it comes from multiple sources. Their marketing is often based on the charm of their personal discovery by the consumer (Louveaux, 1980).

1.5. Honeydew honey

Honeydew honey is produced from the exudation deposited in sticky film on the plants by the secretions of plants (genera *Pinus*, *Abies*, *Castanea*, and *Quercus*, among others) or the excretions of plant-sucking insects mostly from the family *Aphididae* (Pita-Calvo and Vázquez, 2018). The pollen that accompanies the other elements featured in this honey provides information on its geographic origin (Renault et al., 1992). Honeydew honeys have darker color, contain higher values of most physicochemical properties and bioactive compounds and present higher health benefits characteristics compared to floral honeys, which increases its demand commercially (Pita-Calvo and Vázquez, 2018).

In Europe, the main honeydew resources are coniferous and oak forests. All conifers have their parasites, fir (*Abies pectinata*), spruces (*Picea*), pines (*Pinus*), larches (*Larix*), cedars (*Cedrus*), often *Cinara* aphids (each coniferous species has its own *Cinara*). We can also cite *Physokermes piceae*, a parasitic spruce cochineal (*Picea*), which produces honeydew in abundance. The oak (*Quercus*) is parasitized by an aphid, *Lachnus roboris*, its honeydew honey is dark brown (Schweitzer, 2004).

2. Pollen

2.1. Definition

The word comes from Latin which means flower flour, it is the male gamete responsible for the transport of genetic characters from one generation to another. Pollen is in the form of microscopic grains enclosed in the anthers of stamens (Saury, 1981), of variable size and shape. Pollen is usually transported on other flowers either by wind (anemophilous) or by insects (entomophilous) (Marchenay, 1984). It is the main source of nitrogenous food for the bee brood from the larval state to the young adult (Saury, 1981).

Bees ensure the fertilization of 50 to 60% of plant species (Jean-Prost, 1987). Honeybees preferably gather the same kind of flowers at each exit, and therefore collect the same kind of pollen (Fronty, 1997). The production of pollen by the flower is variable in the same plant according to the conditions of the environment, it is a complex phenomenon whose causes are various. According to Rabiet (1989), there are several factors that come into play such as the age of the plant, its vigor and the physiology of the plant.

2.2. Compositions

Pollen is the main source of protein, minerals, fat and many other substances for bees (Herbert, 1992). Pollen is very rich in protein; 100 g of pollen provide as much protein as 7 eggs or 400 g of beef. Pollen richness in amino acids is quantitative but also qualitative (Cherbuliez and Domerego, 2003).

According to Cherbuliez and Domerego (2003) pollen is composed of: 27% carbohydrates, 20% protein: 21 known amino acids, all essential amino acids in interesting proportions: Leucine 9.06%, Lysine 7.70%, Isoleucine 7.00%, Valine 6.91%, Phenylalanine 5.94%, Threonine 5.28%, Methionine 1.17%, Tryptophan less than 1%, 18% of cellulosic substances, 15-18% water, 5% lipids, 5% minerals, vitamins: vitamin A (retinol), vitamin B1 (thiamine), vitamin B2 (riboflavin), vitamin B3/PP (nicotinamide), vitamin B5 (pantothenic acid), vitamin B6 (pyridoxine), vitamin C (ascorbic acid), vitamin E (tocopherol), trace elements, growth hormone, antibiotic substances, 3% of various components not yet identified.

2.3. Usage

According to Chauvin (1968), the consumption of pollen by young honeybees leads to an extension of their lifespan, development of the hypopharyngeal glands and growth of the ovaries of recently hatched honeybees. On the other hand, older honeybees consume little pollen (Jean-Prost, 1987). On the other hand, the action of pollen for the human organism has been studied since 1955. Numerous scientific communications relating to pollen affirm that its beneficial effects are numerous and well-marked (Chauvin, 1968). According to (Jean-Prost, 1987):

- Regulatory action of intestinal functions in patients with chronic constipation, or chronic diarrhea of low origin and resistant to antibiotics.
- In children with anemia, pollen causes a rapid rise in hemoglobin levels in the blood.

- Pollen also brings about a rapid recovery of weight and strength for the convalescent.

2.4. Pollen collection and conservation

To collect pollen for human use, beekeepers use traps of pollen placed at the entrance of the hive, whose mesh size of which should collect 70% of the pollen supplied. The detached balls fall into a drawer. The harvest must be daily because fresh pollen has a very short lifespan and degrades quickly (recovery humidity, fermentations). Pollen is placed on trays, then immediately frozen or dried for ten hours using a stream of hot, dry air. Dried and disinfected with carbon chloride, it can be stored for a long time (Ravazzi, 2003). The pollen harvest is adjusted to the needs of the colony. Over a year, a scrupulous beekeeper leaves enough food for the honeybees. He can harvest between 2 and 4 kg per hive, about 10% of the total harvest (Dreller and Tarp, 2000).

3. Beeswax

3.1. Definition

Beeswax is a fatty substance secreted by the four pairs of wax glands located on the ventral part of the abdomen of workers about two weeks old (Philippe, 1999). When the wax is emitted by the wax glands, it is perfectly white and pure. It is synthesized from honey by chemical reduction of sugars (Louveau, 1980).

According to (Jean-Prost, 1987), this secretion is subject to all the following four factors:

- Presence of honeybees born according to Roesh (1927), from 12 to 18 days and younger according to Lindauer (1963).
- Temperature of 33 to 36 °C of the group of wax caves.
- Copious food, to secrete 1 kg of wax, the workers consume 6 to 12 kg of honey (Bertrand, 1983).
- Need for the colony.

The freshly secreted wax is almost white, it becomes yellow, then very dark brown with age, according to external elements such as the carotenoid pigments of the pollens and the fragments of cocoons in the cells (Philippe, 1999). Beewax has a particular color and smell. Its characteristics are often linked to the plant that produces the raw material (Loiriche, 1984).

3.2. Compositions and properties

Beeswax contains 92-95% pure wax mixed with pollen and propolis (Jiménez et al. 2004). It is a chemically stable fatty substance, with 300 components (Jiménez et al. 2004). Beeswaxes are lipids resulting from the esterification of various alcohols by the corresponding fatty acids. They have great chemical stability and are composed of 72% esters, 13.5% acids, 10.5% hydrocarbons, 1% free alcohols, 0.6% lactones, 0.4% pigments and 2% mineral impurities (Bogdanov, 2009).

3.3. Usage of beeswax

In the form of a cream or ointment, beeswax is used in cosmetology because of the bacteriostatic, emollient, anti-inflammatory and healing properties of several of its constituents (Jean-Prost, 1987). In addition, various industries use it such as pharmaceuticals, armaments, marine, cooking industry.... etc. (Louveaux, 1985).

4. Propolis

4.1. Definition

Propolis, also called bee glue, is the generic of the resinous substance collected by honeybees from various plants (Bankova et al., 2000). Honeybees use it at the entrance to their apiary to protect access, which indicates the Greek etymology "*pro*" which means in front or defense, and "*polis*" the city (Ghisalberti, 1997).

The propolis is a complex of a series of gummy resinous substances, collected by honeybees mainly from plants, trees, tree buds (Kumazawa et al., 2008). Honeybees bring back substances to the hive and they modify them in part by the contribution of some of their own secretions (wax and salivary secretions mainly). Propolis has a balsamic odor and a variable color according to its plant origins, it varies from light yellow to very dark brown almost black (Tosi et al., 2006). Honeybees collect its precious substances from poplar buds, birches, alders, willows, horse chestnut trees, ash trees, spruces and oaks, etc. (Bankova et al., 2000).

Propolis is used by workers to seal the cracks and holes in their hive, or as an antiseptic substance to coat a putrescible foreign body, which they cannot evacuate from the hive. It is also used by bees to coat the cells and in general the entire interior of the hive, which gives it

bactericidal and antiseptic protection (Philippe, 1988). The amount of propolis harvested by bees varies from one breed to another and from one colony to another, a hive can provide up to 300 g per year (Jean-Prost, 1987).

4.2. Compositions and properties of propolis

The composition of propolis varies greatly depending on its source (Donadieu, 1986):

- Wax: the rate varies between 30% and 40%.
- Essential oils: 4.5% but can increase up to 10%
- Aromatic resins: around 50%.
- 5% pollen (the presence of pollen grains in propolis is accidental, just like those found everywhere in the hive).
- 5% of various materials.

4.3. Usage

Propolis is widely used in the food industry, medicine, cosmetology and veterinary medicine (Tosi et al., 2006). In addition, propolis plays an important role in medicine through its innumerable virtues, including its bactericidal effects against a large number of different bacteria, in particular against pathogens such as *Bacillus cereus*, *Bacillus subtilis*, *Staphylococcus aureus* and *Escherichia faocalis* (Ghedira et al., 2009), anti-inflammatories, fungicides, antivirals and cytotoxics (Li et al., 2008).

5. Honeybee venom

5.1. Definition

Honeybee is generally not aggressive, it only stings when it feels threatened. The venom is a colorless liquid, secreted by two glands, one is acid and the other one is alkaline connected to the sting apparatus of the honeybee, located between the fifth and the sixth segment (Ravazzi, 2003).

5.2. Compositions and properties

The main components of honeybee venom are water, formic acid, hydrochloric acid, phosphoric acid, melittin, histamine, apamine. It also contains methionine, cystine, mineral salts and enzymes such as phospholipase and hyaluronidase (Louveaux, 1980).

5.3. Usage

Honeybee venom is as toxic to humans as the venom of the most venomous snakes. It's a question of dose. It is better to leave it to specialists to handle such a dangerous substance, especially when you know that it can cause serious allergies, sometimes fatal. Honeybee venom has a positive effect on rheumatism. Vaccines of honeybee venom are available in eastern countries (Louveau, 1980).

6. Royal jelly

6.1. Definition

Royal jelly has a gelatinous appearance, a light white color and a strong acidic nature (Fronty, 1997), it is secreted by the hypo-pharyngeal and mandibular glands of workers aged 5 to 15 days (Philippe, 1999). It is the food provided to all young larvae, both workers and false bumblebees, during the first three days of their life, while that which will become a queen continues to receive royal jelly throughout its life (Biri, 1986; 1997).

6.2. Compositions

The composition of royal jelly varies according to the nature and age of the larvae to be fed. The grain of pollen contained in royal jelly, like that of honey, can indicate the geographical origin and the harvest season of royal jelly (Jean-Prost, 1987).

Royal jelly contains on average (Philippe, 1999):

- Water by 66%
- Carbohydrates (sugars): 14.5%, we find glucose and fructose for the most part, and in lower proportions of sucrose, maltose and trehalose.
- Lipids (fatty substances) for 4.5% in the form of various fatty acids.
- Proteins (nitrogenous substance): on average 13%, of which a large part in the form of amino acids in the free or combined state (alanine, arginine, aspartic acid ...)
- A very large number of amino acids essential to life that our body cannot synthesize isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine.
- Chemists have been able to highlight the different types of vitamins in the

royal jelly (Vitamins of group B, C, D, Inositol and folic acid).

6.3. Usage

- In the medical field, royal jelly contributes to the balance of basic metabolism.
- Royal jelly helps strengthen the immune system and therefore prevent disease.
- Mixed with honey or pollen, royal jelly action is recognized as very beneficial on ulcers, gastritis and liver problems (Clément, 2000).

Experimental part

Chapter I.

Inventory of

melliferous

plants

1. Materials and Methods

1.1. Study Area

Jijel is a coastal Mediterranean Wilaya located in the northeast of Algeria ($5^{\circ} 25'$ and $6^{\circ} 30'E$ and $36^{\circ} 10'$ and $36^{\circ} 50'N$). In this region, the temperature varies between $20^{\circ}C$ and $35^{\circ}C$ in summer and $5^{\circ}C$ to $15^{\circ}C$ in winter. The rainy season lasts around 06 months, and the average annual precipitation recorded in the wilaya varies from 800 to 1200 mm/year. The plains areas are located north along the coastal strip while the mountainous terrains are south and dominates the Jijelian landforms (82%).

In order to characterize the honey flora of Jijel region, three study stations were chosen representing three different altitudes, which have three different ecosystems, and where there is a remarkable beekeeping activity (more than three apiaries). First, there is a station at 6 m of altitude (A), which is located near the coast of Bni Belaid. The apiary of this area is close to the sand dunes surrounded by an open agricultural land. Moreover, a wetland to the west and a town and maquis to the east surround it. Secondly, there is a station at 70 m of altitude (B), which is an intermediate zone between the coastal and mountainous zone, located in the municipality of Elkennar. It is characterized by a clayey open herbaceous formation (pastureland) and mountains to the south and the town of Elkennar to the north surround it. Finally, a station at 700 m of altitude (C) characterized by forest species, maquis, horticulture and some small plantations (Figure 1).



Figure 4. Locations of the three inventoried stations in Jijel.

1.2. Identification and characterization of the melliferous flora

In order to inventory the plants, an apiary is taken at each station as a starting point and a two-kilometer transect sampling is carried out in four directions for each apiary. The photos of blooming plants were taken to identify them in the laboratory. For this purpose, guides and determination keys were used, namely: Quezel and Santa, 1962-1963; Schauenberg and Ferdinand, 1977; Reisigl and Danesch, 1987; Schönfelder and Schönfelder, 1988; Bayer et al., 1990; Stichmann-Marny, et al., 1997; Bartels, 1998; Blamey and Grey-wdson, 2000; Boucher, 2000; Stichmann and Stichmann-Marny, 2000; Chevallier, 2001; Dietmar, 2004; More and White, 2005; Burnie et al., 2006; Chevallier, 2007; Durcerf, 2007 and Schmidit, 2007.

After specimen's identification, plant specimens that were hard to identify were taken to help us identify them. A melliferous plant was considered once found as a melliferous plant in the bibliography or encountered foraging by bees. In addition, we note the morphological types and the domestication of each species. In order to better characterize the mellific potential of each station (altitude), the most abundant and close plants of each apiary were determined. On the other hand, the flowering schedule of the honey plants was done by a follow-up of each ten day during the year of 2015 then each month in the year of 2016 and the verification of certain species during the years of 2017.

Bibliography and field investigations were used to identify the beekeeping value of each plant. The plant is considered nectariferous if we noticed the extension of proboscis of the foraging bees and polliniferous if we noticed the pollen basket in the posterior feet of foraging bees (Toopchi-Khosroshahi and Lotfalizadeh 2011). On the other hand, bibliography also was use for the plants that were not be able to be investigated. Those bibliography are: Pesson and Louveaux, 1984; Dafni and Dukas, 1986; Lozano et al., 1988; Hidalgo et al., 1990; Mercuri et al., 1991; Zietsman, 1991 ; Nyman, 1992; Verma, 1992; Vidal et al., 2006; Bosch et al., 1997; Kubitzki, 1998; Wickens, 2001; Gaspar et al., 2002 ; Reyes-Agüero et al., 2006; Albano et al., 2009; Chefrour et al., 2009; Makhloufi et al., 2010; Clément, 2011; Pozo, 2011; Rodríguez-Pérez and Traveset, 2011; Leleux, 2012; Song et al., 2012; Yang, et al., 2012; Lopez et al., 2013; Vallés et al., 2013; Zerrouk et al., 2013; Bhalchandra et al., 2014; Isermann and Rooney, 2014; Moisan-de-Serres et al., 2014; Yang et al., 2014; Abrol, 2015a; Abrol, 2015b; Albaba, 2015; Alqarni, 2015 and Rolli et al., 2016.

2. Results and discussion

2.1. Inventoried melliferous plants

The inventory carried out in the Jijel region made it possible to count 296 honey-producing plants (Table 6). The distribution by station of the listed melliferous plants indicates that the 70m zone contains more species than the other zones by 252 (85%) species then the 6m zone by 235 (79%) species and finally the mountain area by 211 (71%) species. On the other hand, Mačukanović-Jocić and Jarić, 2016 reported 197 melliferous plants from Southwestern Vojvodina region in Serbia, Sekine et al., 2013 found 208 melliferous plants in apiaries of the counties of Ubitatã and Nova Aurora in Brazil, Adgaba et al., 2017 indentified 182 mellifeous plants in Saudi Arabic, Bista and Shivakoti, 2001 reported 119 melliferous plants from Dolakha District in Nepal, Toopchi-khosroshahi and Lotfalizadeh, 2011 were able to identify 98 melliferous plants from Kandovan region in Northwest of Iran, Nguemo et al., 2004 and Dongock et al., 2007 found 78 and 88, respectively, melliferous plants in West Cameroon and Taha et al., 2017 reported 110 melliferous plants from Kafrelsheikh province of northern Egypt . Therefore, the region of Jijel showed a good diversity of melliferous plants. The humid tropical zones, the semi-desert zones, the very high mountains and the regions close to the polar circle are not very productive and therefore sparsely populated with bees. Their flora is much less rich in melliferous species than that of temperate regions (Pesson and Louveaux, 1984).

The number of honey plants present in the three altitudes is 186 (63%) species. However, the species common between the different altitudes are 203 (67%) species between the 70m zone and the high-altitude zone, 196 (66%) species between the low altitude zone and the 70m zone, and 187 (63%) species between the 6m zone and the high-altitude zone (Figure 2). These results seem compatible with the fact that the 70m zone is a transitional ecosystem to a mountain ecosystem, which makes it have more similarities in species with mountain zone. On the other hand, 38 species were found only at 70m station (*Acacia saligna* (Labill.) H.L.Wendl., *Bupleurum lancifolium* Hornem., *Carpobrotus edulis* (L.) N.E.BR., *Cestrum nocturnum* L., *Carlina racemosa* L.....), 37 species were found only at 6m station (*Glaucium flavum* Crantz , *Cakile maritima* Scop., *Dipsacus fullonum* L., *Echinophora spinosa* L., *Echinops spinosus* L., *Eryngium maritimum* L.....) and 6 species were found only at 700m station (*Castanea sativa* Mill., *Centranthus ruber* (L.) DC., *Prunus avium* (L.) L. *Prunus dulcis* (Mill.) D.A.Webb, *Silene latifolia* Poir., *Simethis mattiazzii* (Vand.) G.López& Jarvis) (Figure 1). In addition, *Lotus Tetragonolobus* L. was only

found during the year of 2015 at the 70m zone and not in 2016 and 2017, even though it is an endemic species in the Mediterranean region that develops in hilly pastures up to 1200 m (Vargiu and Spanu, 2016). Among the 38 species only present in 70m zone there are 14 planted plants, which means that these plants can be found in the other zones. Therefore, the 6m zone had more unique flora compared to the others.

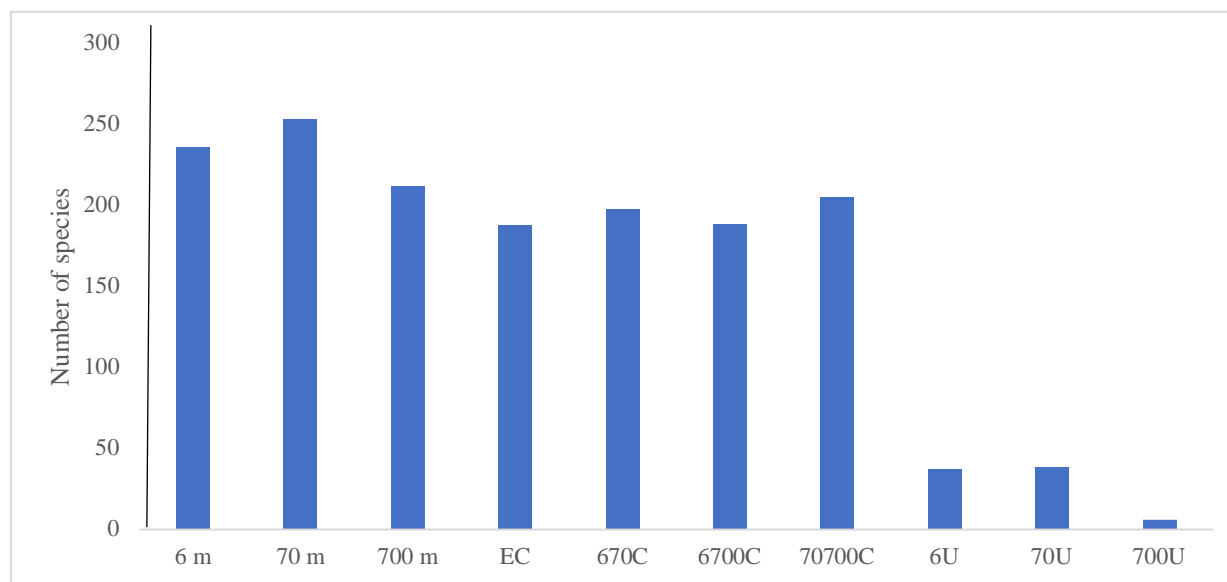


Figure 5. Distribution of melliferous plants at the three stations. EC= common species in the three stations, 670C= common species between 6m and 70m, 6700C= common species between 6m and 700 m, 70700C= common species between 70 m and 700 m, 6U= species present only in 6m, 70U= species present only in 70 m, 700U= species present only in 700 m.

The total number of identified plants in the Jijel region has reached 296 species. This floristic richness in honey plants is mainly due to the presence of several plant formations, ranging from sand dunes to forested areas, through agricultural areas and pastureland. Knowing that this floristic richness is relative for each station. The 70m station contains more species than the other stations because of the intermediate position between the coastal and forest ecosystems, which gives it a great diversity of flora. In addition, the low altitude station contains more species than the high altitude station due to the presence of several vegetation formations (sand dunes, riparian plants, crop fields, horticulture, etc.) compared to the high altitude station, which is a homogeneous forestry with some fruit trees and some small crops. The presence of 186 common species in the three stations means that 63% of the honey flora is ubiquitous in this region.

Table 6. Inventory, beekeeping value, period of flowering, morphological types and domestication of melliferous plants of Jijel.

Melliferous plants	Family	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Acacia karroo</i> Hayne	<i>Fabaceae</i>	x	x	J-O	st	PL	x	x	
<i>Acacia saligna</i> (Labill.) H.L. Wendl.	<i>Fabaceae</i>	x	x	M-MY	st	PL		x	
<i>Acanthus molis</i> L.	<i>Acanthaceae</i>	x	x	A-J	h	L	x	x	x
<i>Alisma lanceolatum</i> With.	<i>Alismaceae</i>	x		J-S	h	L	x	x	
<i>Allium cepa</i> L.	<i>Liliaceae</i>	x	x	M-MY	h	PL	x	x	x
<i>Allium roseum</i> L.	<i>Liliaceae</i>	x		A-MY	h	L	x	x	x
<i>Allium sativum</i> L.	<i>Liliaceae</i>	x	x	A-MY	h	PL	x	x	x
<i>Allium triquetrum</i> L.	<i>Liliaceae</i>	x	x	M-A	h	L	x	x	x
<i>Alnus glutinosa</i> (L.) Gaertn.	<i>Betulaceae</i>		x	J-A	t	L	x		
<i>Andryala integrifolia</i> L.	<i>Asteraceae</i>		x	A-N	h	L	x	x	x
<i>Anthemis arvensis</i> L.	<i>Asteraceae</i>	x	x	D-J	h	L	x	x	x
<i>Anthemis mixta</i> L.	<i>Asteraceae</i>		x	MY-N	h	L	x	x	x
<i>Anthemis nobilis</i> L.	<i>Asteraceae</i>		x	M-MY	h	L	x	x	x
<i>Apium graveolens</i> L.	<i>Apiaceae</i>	x	x	J-O	h	PL	x	x	x
<i>Arbutus unedo</i> L.	<i>Ericaceae</i>	x		S-N	sh	L	x	x	x
<i>Asparagus acutifolius</i> L.	<i>Asparagaceae</i>	x	x	A-J	l	L	x	x	x
<i>Asphodelus microcarpus</i> Viv.	<i>Liliaceae</i>	x		M-MY	h	L	x	x	x
<i>Bartsia trixago</i> L.	<i>Scrophulariaceae</i>	x		M-MY	h	L		x	x
<i>Bartsia viscosa</i> L.	<i>Scrophulariaceae</i>	x		A-MY	h	L		x	x
<i>Bellevia romana</i> (L.) Rchb.	<i>Hyacinthaceae</i>	x	x	M-A	h	L		x	x
<i>Bellis annua</i> L.	<i>Asteraceae</i>		x	N-MY	h	L	x	x	x
<i>Bellis sylvestris</i> Cyr.	<i>Asteraceae</i>	x	x	OC-J	h	L	x	x	x
<i>Blackstonia perfoliata</i> (L.) Huds.	<i>Gentianaceae</i>		x	M-J	h	L	x	x	x
<i>Borago officinalis</i> L.	<i>Boraginaceae</i>	x		M-MY	h	L	x	x	x
<i>Bougainvillea glabra</i> Choisy	<i>Nyctaginaceae</i>	x	x	D-N	sh	PL		x	
<i>Bryonia cretica</i> L.	<i>Cucurbitaceae</i>	x	x	M-MY	l	L	x	x	x
<i>Bupleurum lancifolium</i> Hornem.	<i>Apiaceae</i>	x	x	A-MY	h	L		x	
<i>Cakile maritima</i> Scop.	<i>Brassicaceae</i>	x	x	MY-S	h	L	x		
<i>Calendula arvensis</i> L.	<i>Asteraceae</i>		x	A-J	h	L	x	x	x
<i>Calicotome spinosa</i> (L.) Link.	<i>Fabaceae</i>	x	x	M-MY	sh	L	x	x	x
<i>Campanula dichotoma</i> L.	<i>Campanulaceae</i>	x	x	A-J	h	L	x	x	x
<i>Campanula</i> sp.	<i>Campanulaceae</i>	x	x	A-J	h	L		x	
<i>Capsella bursa-pastoris</i> (L.) Medik.	<i>Brassicaceae</i>	x		M-A/N-D	h	L	x	x	x
<i>Capsicum annuum</i> L.	<i>Solanaceae</i>		x	MY-N	h	PL	x	x	x
<i>Carduncellus caeruleus</i> (L.) C. Presl	<i>Asteraceae</i>	x	x	A-J	h	L		x	x
<i>Carlina corymbosa</i> L.	<i>Asteraceae</i>	x	x	J-O	h	L	x		x
<i>Carlina gummifera</i> (L.) Less.	<i>Asteraceae</i>	x	x	JU-S	h	L		x	x
<i>Carlina racemosa</i> L.	<i>Asteraceae</i>	x	x	O-OC	h	L		x	
<i>Carpobrotus edulis</i> (L.) N.E.BR.	<i>Aizoaceae</i>	x	x	MY-JU	h	L		x	
<i>Castanea sativa</i> Mill.	<i>Fagaceae</i>	x	x	MY-JU	t	L			x
<i>Catharanthus roseus</i> (L.) G. Don.	<i>Apocynaceae</i>	x		A-D	h	PL	x	x	x
<i>Celtis australis</i> L.	<i>Ulmaceae</i>	x	x	F-A	t	L	x	x	x
<i>Centaurea calcitrapa</i> L.	<i>Asteraceae</i>	x	x	MY-JU	h	L	x	x	x
<i>Centaurea</i> sp.	<i>Asteraceae</i>	x	x	A-O	h	L		x	
<i>Centaurea sphaerocephala</i> L.	<i>Asteraceae</i>	x	x	A-OC	h	L	x	x	x
<i>Centaurium erythraea</i> Rafn	<i>Gentianaceae</i>	x	x	MY-J	h	L	x	x	x
<i>Centranthus ruber</i> (L.) DC.	<i>Caryophyllaceae</i>	x		MY-S	h	L			x
<i>Cestrum nocturnum</i> L.	<i>Solanaceae</i>	x		MY-N	st	PL		x	

Melliferous plants	Family	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Chondrilla juncea</i> L.	Asteraceae	x	x	JU-S	h	L	x	x	
<i>Chrysanthemum coronarium</i> L.	Asteraceae		x	M-MY	h	L		x	x
<i>Chrysanthemum segetum</i> L.	Asteraceae		x	MY-O	h	L	x		
<i>Chrysanthemum</i> sp.	Asteraceae		x	OC-F	h	L		x	
<i>Cichorium intybus</i> L.	Asteraceae	x	x	A-O	h	L	x	x	x
<i>Cistus monspeliensis</i> L.	Cistaceae	x	x	A-MY	sh	L	x	x	x
<i>Cistus salviaefolius</i> L.	Cistaceae	x	x	A-J	sh	L	x	x	x
<i>Citrullus lanatus</i> (Thunb.) Matsum. & Nakai	Cucurbitaceae	x	x	MY-O	l	PL	x		
<i>Citrus aurantium</i> L.	Rutaceae	x	x	A-MY	st	PL		x	
<i>Citrus limon</i> (L.) Burm.f.	Rutaceae	x	x	D-N	st	PL	x	x	x
<i>Citrus sinensis</i> (L.) Osbeck	Rutaceae	x	x	A-MY	st	PL	x	x	x
<i>Convolvulus althaeoides</i> L.	Convolvulaceae	x	x	A-MY	h	L	x	x	x
<i>Convolvulus arvensis</i> L.	Convolvulaceae	x	x	A-D	l	L	x	x	x
<i>Convolvulus elegantissimus</i> Mill.	Convolvulaceae	x	x	A-J	h	L		x	x
<i>Convolvulus sabatius</i> Viv.	Convolvulaceae	x	x	MY-J	h	L		x	
<i>Convolvulus sepium</i> L.	Convolvulaceae	x	x	M-J	l	L	x	x	x
<i>Coriandrum sativum</i> L.	Apiaceae	x	x	A-O	h	PL	x	x	x
<i>Crataegus monogyna</i> Jacq.	Rosaceae	x	x	M-MY	sh	L	x	x	x
<i>Cucumis melo</i> L.	Cucurbitaceae	x	x	J-S	l	PL	x		
<i>Cucumis sativus</i> L.	Cucurbitaceae	x	x	F-MY	l	PL	x	x	x
<i>Cucurbita pepo</i> L.	Cucurbitaceae	x	x	A-D	l	PL	x	x	x
<i>Cupressus sempervirens</i> L.	Cupressaceae		x	M-MY	t	PL		x	
<i>Cydonia oblonga</i> Mill.	Rosaceae	x	x	A-J	st	PL		x	x
<i>Cynara scolymus</i> L.	Asteraceae	x		M-MY	h	PL	x	x	x
<i>Cynoglossum cheirifolium</i> L.	Boraginaceae	x		M-A	h	L	x	x	x
<i>Cynoglossum creticum</i> Miller	Boraginaceae	x		M-MY	h	L	x	x	x
<i>Cytisus triflorus</i> Lam.	Fabaceae	x		A-MY	sh	L	x	x	x
<i>Daphne gnidium</i> L.	Thymelaeaceae	x	x	J-D	sh	L	x	x	x
<i>Datura stramonium</i> L.	Solanaceae	x	x	A-N	h	L	x	x	x
<i>Daucus carota</i> L.	Apiaceae	x	x	M-N	h	L	x	x	x
<i>Dipsacus fullonum</i> L.	Dipsacaceae	x	x	MY-O	h	L	x		
<i>Echinophora spinosa</i> L.	Apiaceae	x		JU-OC	h	L	x		
<i>Echinops spinosus</i> L.	Asteraceae	x	x	MY-JU	h	L	x		
<i>Echium plantagineum</i> L.	Boraginaceae	x	x	A-MY	h	L	x	x	x
<i>Echium vulgare</i> L.	Boraginaceae	x	x	M-O	h	L	x	x	x
<i>Erica arborea</i> L.	Ericaceae	x	x	M-MY	sh	L	x	x	x
<i>Eriobotrya japonica</i> (Thunb.) Lindl.	Rosaceae	x	x	N-J	st	PL	x	x	x
<i>Erodium cicutarium</i> (L.) L'Hér.	Geraniaceae	x	x	J-MY	h	L	x	x	x
<i>Erodium</i> sp.	Geraniaceae	x	x	MY-J	h	L		x	
<i>Eryngium bourgatii</i> Gouan	Apiaceae	x	x	O-N	h	L	x	x	x
<i>Eryngium maritimum</i> L.	Apiaceae	x	x	J-S	h	L	x		
<i>Eucalyptus</i> sp.	Myrtaceae	x	x	MY-O	t	PL	x	x	x
<i>Euphorbia biumbellata</i> Poir.	Euphorbiaceae	x		A-J	h	L	x	x	x
<i>Euphorbia helioscopia</i> L.	Euphorbiaceae	x	x	A-O	h	L	x	x	x
<i>Fedia cornucopiae</i> (L.) Gaertn.	Valerianaceae	x	x	F-MY	h	L	x	x	x
<i>Foeniculum vulgare</i> L.	Apiaceae	x	x	MY-S	h	PL	x	x	x
<i>Fragaria × ananassa</i> (Weston) Duchesne ex Rozier	Rosaceae	x	x	JA-J	h	PL	x	x	x
<i>Fraxinus angustifolia</i> Vahl	Oleaceae	x		A-MY	t	L	x	x	x
<i>Fumaria capreolata</i> L.	Papaveraceae	x	x	J-MY	h	L	x	x	x
<i>Fumaria officinalis</i> L.	Papaveraceae	x	x	A-J	h	L	x	x	x
<i>Galactites tomentosa</i> Moench	Asteraceae	x	x	M-JU	h	L	x	x	x

Melliferous plants	Family	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Galium corrudifolium</i> Vill.	Rubiaceae	x	x	A-J	h	L	x	x	x
<i>Galium palustre</i> L.	Rubiaceae	x	x	MY-O	h	L	x	x	x
<i>Genista tricuspidata</i> Desf.	Fabaceae	x	x	M-MY	sh	L		x	x
<i>Genista ulicina</i> Spach	Fabaceae	x	x	A-MY	sh	L		x	x
<i>Geranium dissectum</i> L.	Geraniaceae	x	x	A-MY	h	L	x	x	x
<i>Geranium pyrenaicum</i> Burm.f.	Geraniaceae	x	x	J-J	h	L	x	x	x
<i>Geranium robertianum</i> L.	Geraniaceae	x	x	A-MY	h	L	x	x	x
<i>Gladiolus communis</i> subsp. <i>byzantinus</i> (Mill.) Douin	Iridaceae	x	x	A-J	h	L	x	x	x
<i>Gladiolus</i> sp.	Iridaceae	x	x	M-MY	h	L		x	
<i>Glaucium flavum</i> Crantz	Papaveraceae	x	x	MY-JU	h	L	x		
<i>Grewia occidentalis</i> L.	Tiliaceae	x	x	J-D	st	PL		x	
<i>Hedypnois cretica</i> (L.) Dum.Cours.	Asteraceae	x	x	A-MY	h	L	x		
<i>Hedysarum coronarium</i> L.	Fabaceae	x	x	A-J	h	L	x	x	x
<i>Helianthus annuus</i> L.	Asteraceae	x	x	JU-OC	h	PL	x	x	x
<i>Heliotropium europaeum</i> L.	Boraginaceae	x		MY-N	h	L	x	x	x
<i>Helosciadium crassipes</i> W.D.J.Koch	Apiaceae		x	A-J	h	L	x		
<i>Hibiscus rosa-sinensis</i> L.	Malvaceae	x		A-J	sh	PL	x	x	
<i>Hyoseris radiata</i> L.	Asteraceae		x	A-JU	h	L	x		
<i>Hypericum perforatum</i> L.	Hypericaceae	x	x	M-MY	h	L	x	x	x
<i>Inula viscosa</i> (L.) Aiton	Asteraceae	x	x	S-N	h	L	x	x	x
<i>Ipomoea tricolor</i> Cav.	Convolvulaceae	x	x	A-J	l	PL		x	
<i>Iris florentina</i> L.	Iridaceae	x	x	M-MY	h	L		x	
<i>Iris foetidissima</i> L.	Iridaceae	x	x	M-J	h	L	x		
<i>Iris pseudacorus</i> L.	Iridaceae	x	x	M-J	h	L	x		
<i>Iris sisyrinchium</i> L.	Iridaceae	x		M-MY	h	L		x	x
<i>Jacaranda mimosifolia</i> D.Don	Bignoniaceae	x	x	A-MY	t	PL		x	
<i>Juglans regia</i> L.	Juglandaceae		x	A-J	t	PL	x	x	x
<i>Lactuca sativa</i> L.	Asteraceae	x	x	A-MY	h	PL	x	x	x
<i>Lamium amplexicaule</i> L.	Lamiaceae	x		A-J	h	L	x	x	x
<i>Lantana camara</i> L.	Verbenaceae	x		A-D	sh	PL	x	x	x
<i>Lathyrus latifolius</i> L.	Fabaceae	x	x	A-MY	l	L	x	x	x
<i>Lavandula stoechas</i> L.	Lamiaceae	x	x	M-MY	ssh	L	x	x	x
<i>Lavatera cretica</i> L.	Malvaceae	x	x	MY-J	h	L	x	x	x
<i>Lavatera olbia</i> L.	Malvaceae	x	x	A-JU	ssh	L	x	x	x
<i>Lavatera</i> sp.	Malvaceae	x	x	A-JU	h	L		x	
<i>Lavatera trimestris</i> L.	Malvaceae	x	x	A-J	h	L		x	x
<i>Ligustrum lucidum</i> W.T.Aiton	Oleaceae	x	x	A-J	st	PL	x	x	x
<i>Linaria</i> sp.	Scrophulariaceae	x	x	N-F	h	L		x	
<i>Linum usitatissimum</i> L.	Linaceae	x		M-MY	h	L	x	x	x
<i>Lippia citriodora</i> Kunth	Verbenaceae	x	x	A-D	sh	PL	x	x	x
<i>Lobularia maritima</i> (L.) Desv.	Brassicaceae		x	S-J	h	L	x	x	x
<i>Lonicera</i> sp.	Caprifoliaceae	x	x	A-J	sh	L		x	x
<i>Lotus cytisoides</i> L.	Fabaceae	x	x	M-MY	h	L	x	x	x
<i>Lotus edulis</i> L.	Fabaceae	x	x	F-J	h	L	x	x	x
<i>Lotus hispidus</i> Desf.	Fabaceae	x	x	A-J	h	L	x	x	x
<i>Lotus Ornithopodioides</i> L.	Fabaceae	x	x	M-J	h	L	x	x	x
<i>Lotus Tetragonolobus</i> L.	Fabaceae	x	x	M-A	h	L		x	
<i>Ludwigia peploides</i> (Kunth) P.H.Raven	Onagraceae	x	x	J-S	h	L	x		
<i>Luffa aegyptiaca</i> L.	Cucurbitaceae	x	x	J-D	l	PL	x	x	x
<i>Lupinus angustifolius</i> L.	Fabaceae	x	x	M-MY	h	L	x	x	

Melliferous plants	Family	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Lupinus luteus</i> L.	<i>Fabaceae</i>	x	x	M-MY	h	L	x	x	x
<i>Lythrum hyssopifolia</i> L.	<i>Lythraceae</i>	x	x	MY-O	h	L	x	x	
<i>Lythrum junceum</i> L.	<i>Lythraceae</i>	x	x	A-J	h	L	x	x	x
<i>Lythrum salicaria</i> L.	<i>Lythraceae</i>	x	x	J-S	h	L	x		
<i>Malus pumila</i> Mill.	<i>Rosaceae</i>	x	x	M-MY	t	PL	x	x	x
<i>Malva neglecta</i> Wallr.	<i>Malvaceae</i>		x	A-MY	h	L	x	x	x
<i>Malva sylvestris</i> L.	<i>Malvaceae</i>	x	x	A-OC	h	L	x	x	x
<i>Medicago marina</i> L.	<i>Fabaceae</i>	x	x	A-J	h	L	x		
<i>Medicago polymorpha</i> L.	<i>Fabaceae</i>	x	x	A-J	h	L	x	x	x
<i>Medicago sativa</i> L.	<i>Fabaceae</i>	x	x	A-J	h	L		x	
<i>Melia azedarach</i> L.	<i>Meliaceae</i>	x	x	A-D	t	PL		x	
<i>Melilotus albus</i> Medik.	<i>Fabaceae</i>	x	x	M-MY	h	L		x	
<i>Melilotus officinalis</i> (L.) Lam.	<i>Fabaceae</i>	x	x	A-J	h	L	x	x	x
<i>Melilotus sulcata</i> Desf.	<i>Fabaceae</i>	x	x	MY-J	h	L	x	x	x
<i>Mentha aquatica</i> L.	<i>Lamiaceae</i>	x	x	J-O	h	L	x	x	x
<i>Mentha pulegium</i> L.	<i>Lamiaceae</i>	x	x	MY-OC	h	L	x	x	x
<i>Mentha rotundifolia</i> L.	<i>Lamiaceae</i>	x	x	MY-JU	h	L	x	x	x
<i>Mirabilis jalapa</i> L.	<i>Nyctaginaceae</i>	x	x	A-D	h	PL	x	x	x
<i>Musa</i> sp.	<i>Musaceae</i>	x	x	J-D	h	PL	x	x	x
<i>Myrtus communis</i> L.	<i>Myrtaceae</i>	x	x	A-MY	sh	L	x	x	x
<i>Narcissus tazetta</i> L.	<i>Amaryllidaceae</i>		x	F-A	h	L	x		
<i>Nigella damascena</i> L.	<i>Ranunculaceae</i>		x	A-J	h	L	x	x	x
<i>Oenanthe fistulosa</i> L.	<i>Apiaceae</i>	x	x	MY-O	h	L	x		
<i>Olea</i> sp.	<i>Oleaceae</i>		x	A-JU	t	PL	x	x	x
<i>Ononis hispidula</i> Desf.	<i>Fabaceae</i>	x		MY-JU	h	L		x	x
<i>Ononis repens</i> L.	<i>Fabaceae</i>	x		J-O	h	L	x		
<i>Ononis variegata</i> L.	<i>Fabaceae</i>	x	x	A-J	h	L	x		
<i>Opuntia ficus-indica</i> (L.) Mill.	<i>Cactaceae</i>	x	x	A-MY	st	PL	x	x	x
<i>Origanum</i> sp.	<i>Lamiaceae</i>	x	x	JU-D	h	PL		x	
<i>Ornithogalum narbonense</i> L.	<i>Liliaceae</i>	x	x	A-J	h	L	x	x	x
<i>Oxalis corniculata</i> L.	<i>Oxalidaceae</i>		x	M-J	h	L	x	x	x
<i>Oxalis pes-caprae</i> L.	<i>Oxalidaceae</i>	x	x	N-MY	h	L	x	x	x
<i>Pallenis spinosa</i> (L.) Cass.	<i>Asteraceae</i>		x	M-J	h	L	x	x	x
<i>Pancratium maritimum</i> L.	<i>Amaryllidaceae</i>		x	J-S	h	L	x		
<i>Papaver dubium</i> L.	<i>Papaveraceae</i>		x	MY-J	h	L	x	x	x
<i>Papaver rhoeas</i> L.	<i>Papaveraceae</i>		x	M-MY	h	L	x	x	x
<i>Parthenocissus quinquefolia</i> (L.) Planch.	<i>Vitaceae</i>	x	x	MY-S	l	PL	x	x	x
<i>Pelargonium graveolens</i> L'Hér.	<i>Geraniaceae</i>	x		A-J	ssh	PL	x	x	x
<i>Pelargonium</i> sp.	<i>Geraniaceae</i>	x		J-D	h	PL		x	
<i>Petroselinum crispum</i> (Mill.) Fuss	<i>Apiaceae</i>	x	x	M-O	h	PL	x	x	x
<i>Phaseolus vulgaris</i> L.	<i>Fabaceae</i>	x	x	A-MY/ N-D	h	PL	x	x	x
<i>Phoenix dactylifera</i> L.	<i>Arecaceae</i>		x	M-A	t	PL	x	x	x
<i>Phyla nodiflora</i> (L.) Greene	<i>Verbenaceae</i>	x		M-S	h	L	x		
<i>Picris echioides</i> L.	<i>Asteraceae</i>	x	x	J-N	h	L	x	x	x
<i>Pinus maritima</i> Lam.	<i>Pinaceae</i>		x	A-MY	t	L		x	
<i>Pistacia lentiscus</i> L.	<i>Anacardiaceae</i>		x	M-MY	sh	L	x	x	x
<i>Pisum sativum</i> L.	<i>Fabaceae</i>	x	x	N-M	h	PL	x	x	x
<i>Populus alba</i> L.	<i>Salicaceae</i>		x	J-A	t	L	x	x	x
<i>Populus nigra</i> L.	<i>Salicaceae</i>		x	F-A	t	L	x	x	x
<i>Portulaca oleracea</i> L.	<i>Portulacaceae</i>		x	JU-OC	h	L	x	x	x

Melliferous plants	Family	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Prasium majus</i> L.	Lamiaceae	x			sh	L	x		
<i>Prunella vulgaris</i> L.	Lamiaceae	x	x	A-J	h	L	x	x	
<i>Prunus armeniaca</i> L.	Rosaceae	x	x	F-M	st	PL	x	x	x
<i>Prunus avium</i> (L.) L.	Rosaceae	x	x	M-A	t	PL			x
<i>Prunus domestica</i> L.	Rosaceae	x	x	M-A	st	PL	x	x	x
<i>Prunus dulcis</i> (Mill.) D.A.Webb	Rosaceae	x	x	F-A	st	PL			x
<i>Prunus persica</i> (L.) Batsch	Rosaceae	x	x	M-A	st	PL	x	x	x
<i>Prunus spinosa</i> L.	Rosaceae	x	x	J-M	sh	L	x	x	x
<i>Pseudorhiza pumila</i> (L.) Grande	Apiaceae		x	M-J	h	L	x		
<i>Pulicaria dysenterica</i> (L.) Bernh.	Asteraceae	x		O-OC	h	L	x	x	x
<i>Punica granatum</i> L.	Punicaceae	x	x	MY-O	sh	PL	x	x	x
<i>Pyrus communis</i> L.	Rosaceae	x	x	M-A	t	PL	x	x	x
<i>Quercus suber</i> L.	Fagaceae		x	A-MY	t	L	x	x	x
<i>Ranunculus acris</i> Jordan	Ranunculaceae	x	x	M-MY	h	L		x	
<i>Ranunculus baudotii</i> Godr.	Ranunculaceae	x	x	A-O	h	L	x		
<i>Ranunculus bulbosus</i> L.	Ranunculaceae	x	x	A-MY	h	L	x	x	x
<i>Ranunculus muricatus</i> L.	Ranunculaceae	x	x	A-MY	h	L	x	x	x
<i>Ranunculus ophioglossifolius</i> Vill.	Ranunculaceae	x	x	A-JU	h	L	x		
<i>Ranunculus sardous</i> Crantz	Ranunculaceae	x	x	A-J	h	L		x	
<i>Ranunculus sceleratus</i> L.	Ranunculaceae	x	x	A-OC	h	L	x		
<i>Ranunculus trichophyllus</i> Chaix	Ranunculaceae	x	x	A-O	h	L	x		
<i>Raphanus raphanistrum</i> L.	Brassicaceae	x	x	M-MY	h	L	x	x	x
<i>Reseda alba</i> L.	Resedaceae	x	x	A-J	h	L	x	x	x
<i>Retama monosperma</i> (L.) Boiss.	Fabaceae	x	x	M-MY	sh	L	x		
<i>Ricinus communis</i> L.	Euphorbiaceae		x	A-D	sh	L	x	x	x
<i>Robinia pseudacacia</i> L.	Fabaceae	x	x	A-J	t	PL		x	
<i>Rosa sempervirens</i> L.	Rosaceae	x	x	J-J	sh	L	x	x	x
<i>Rosa</i> sp.	Rosaceae	x	x	J-D	sh	PL	x	x	x
<i>Rosmarinus officinalis</i> L.	Lamiaceae	x	x	J-D	ssh	L	x	x	x
<i>Rubus ulmifolius</i> Schott	Rosaceae	x	x	A-F	sh	L	x	x	x
<i>Rudbeckia</i> sp.	Asteraceae		x	JU-OC	h	PL		x	
<i>Ruta chalepensis</i> L.	Rutaceae	x		A-J	h	L	x	x	
<i>Salvia</i> sp.	Lamiaceae	x		A-MY	h	L		x	
<i>Salvia verbenaca</i> (L.) Briquet	Lamiaceae	x		F-A	h	L	x	x	x
<i>Sambucus</i> sp.	Caprifoliaceae	x	x	JU-O	sh	L		x	
<i>Scabiosa maritima</i> L.	Dipsacaceae	x	x	MY-N	h	L	x	x	x
<i>Schinus molle</i> L.	Anacardiaceae	x	x	A-S	t	PL	x	x	
<i>Scilla maritima</i> L.	Liliaceae	x	x	O-OC	h	L	x	x	x
<i>Scilla peruviana</i> L.	Liliaceae	x		A-J	h	L	x	x	x
<i>Scolymus grandiflorus</i> Desf.	Asteraceae		x	A-J	h	L		x	
<i>Scolymus hispanicus</i> L.	Asteraceae		x	MY-OC	h	L	x	x	x
<i>Scolymus maculatus</i> L.	Asteraceae		x	MY-JU	h	L	x	x	x
<i>Senecio jacobaea</i> L.	Asteraceae	x	x	N-J	h	L	x	x	x
<i>Senecio leucanthemifolius</i> Poir.	Asteraceae	x	x	F-MY	h	L	x		
<i>Senecio vulgaris</i> L.	Asteraceae	x	x	OC-MY	h	L	x	x	x
<i>Silene colorata</i> L.	Caryophyllaceae	x		A-MY	h	L	x	x	x
<i>Silene dioica</i> (L.) Clairv.	Caryophyllaceae	x	x	A-MY	h	L	x	x	x
<i>Silene gallica</i> L.	Caryophyllaceae	x		N-J	h	L	x	x	x
<i>Silene latifolia</i> Poir.	Caryophyllaceae	x	x	A-J	h	L			x
<i>Silene nicaeensis</i> All.	Caryophyllaceae	x	x	A-J	h	L	x	x	
<i>Silene</i> sp.	Caryophyllaceae		x	J-M	h	L		x	
<i>Silene vulgaris</i> (Moench) Garcke	Caryophyllaceae	x	x	M-MY	h	L	x	x	x

Melliferous plants	Familly	N	P	FP	MT	C	St 6m	St 70m	St 700m
<i>Silybum marianum</i> (L.) Gaertn.	<i>Asteraceae</i>	x	x	A-MY	h	L	x	x	x
<i>Simethis mattiazzii</i> (Vand.) G.López& Jarvis	<i>Xanthorrhoeaceae</i>	x	x	M-JU	h	L			x
<i>Sinapis arvensis</i> L.	<i>Brassicaceae</i>	x	x	M-MY	h	L	x	x	x
<i>Sisymbrium officinale</i> (L.) Scop.	<i>Brassicaceae</i>	x	x	A-J	h	L	x	x	x
<i>Smilax aspera</i> L.	<i>Smilacaceae</i>	x	x	OC-N	l	L	x	x	x
<i>Smyrniolum olusatrum</i> L.	<i>Apiaceae</i>	x	x	M-MY	h	L		x	x
<i>Solanum dulcamara</i> L.	<i>Solanaceae</i>	x		A-MY	l	L		x	x
<i>Solanum lycopersicum</i> L.	<i>Solanaceae</i>	x	x	MY-OC	h	PL	x	x	x
<i>Solanum melongena</i> L.	<i>Solanaceae</i>	x	x	JU-N	h	PL	x	x	x
<i>Solanum nigrum</i> L.	<i>Solanaceae</i>		x	M-D	h	L	x	x	x
<i>Solanum pseudocapsicum</i> L.	<i>Solanaceae</i>		x	MY-J	ssh	PL	x	x	x
<i>Solanum sodomaeum</i> L.	<i>Solanaceae</i>		x	MY-O	ssh	L	x		
<i>Solanum tuberosum</i> L.	<i>Solanaceae</i>		x	F-MY	h	PL	x	x	x
<i>Sonchus oleraceus</i> L.	<i>Asteraceae</i>	x	x	J-D	h	L	x	x	x
<i>Stachys marrubifolia</i> Viv.	<i>Lamiaceae</i>	x	x	MY-O	h	L	x		
<i>Stachys ocymastrum</i> (L.) Briq.	<i>Lamiaceae</i>	x	x	A-J	h	L	x	x	x
<i>Stachys arvensis</i> (L.) L.	<i>Lamiaceae</i>	x	x	MY-O	h	L	x	x	x
<i>Tamarix galica</i> L.	<i>Tamaricaceae</i>	x	x	M-MY	sh	L	x	x	x
<i>Tammus communis</i> L.	<i>Dioscoreaceae</i>	x	x	M-MY	l	L	x	x	x
<i>Taraxacum officinalis</i> Weber	<i>Asteraceae</i>	x	x	M-MY	h	L	x	x	x
<i>Thapsia villosa</i> L.	<i>Apiaceae</i>	x	x	A-J	h	L		x	
<i>Tolpis barbata</i> (L.) Gaertn.	<i>Asteraceae</i>		x	A-J	h	L	x	x	x
<i>Torilis arvensis</i> (Huds.) Link	<i>Apiaceae</i>	x	x	A-S	h	L	x	x	x
<i>Trifolium angustifolium</i> L.	<i>Fabaceae</i>	x	x	MY-J	h	L	x	x	x
<i>Trifolium arvense</i> L.	<i>Fabaceae</i>	x	x	MY-J	h	L		x	
<i>Trifolium campestre</i> Schreb.	<i>Fabaceae</i>	x		A-J	h	L	x	x	x
<i>Trifolium cherleri</i> L.	<i>Fabaceae</i>	x	x	A-J	h	L		x	x
<i>Trifolium fragiferum</i> L.	<i>Fabaceae</i>	x		MY-O	h	L	x	x	x
<i>Trifolium pratense</i> L.	<i>Fabaceae</i>	x	x	A-MY	h	L	x	x	x
<i>Trifolium repens</i> L.	<i>Fabaceae</i>	x	x	M-J	h	L	x	x	x
<i>Trifolium resupinatum</i> L.	<i>Fabaceae</i>	x	x	A-O	h	L	x	x	x
<i>Ulmus minor</i> Mill.	<i>Ulmaceae</i>	x	x	F-A	t	L	x		
<i>Verbascum sinuatum</i> L.	<i>Scrophulariaceae</i>	x	x	MY-N	h	L	x	x	x
<i>Verbena officinalis</i> L.	<i>Verbenaceae</i>	x		A-D	h	L	x	x	x
<i>Veronica persica</i> Poir.	<i>Scrophulariaceae</i>	x	x	D-MY	h	L	x	x	x
<i>Veronica</i> sp.	<i>Scrophulariaceae</i>	x	x	MY-J	h	L		x	
<i>Vicia faba</i> L.	<i>Fabaceae</i>	x	x	F-A	h	PL	x	x	x
<i>Vicia sativa</i> L.	<i>Fabaceae</i>	x		A-J	h	L	x	x	x
<i>Vinca difformis</i> Pourr.	<i>Apocynaceae</i>	x		N-MY	ssh	L	x	x	x
<i>Visnaga daucooides</i> Gaertn.	<i>Apiaceae</i>		x	JU-OC	h	L	x		
<i>Vitis vinifera</i> L.	<i>Vitaceae</i>	x	x	A-MY	l	PL	x	x	x
<i>Xanthium strumarium</i> L.	<i>Asteraceae</i>		x	JU-OC	h	L	x	x	x
<i>Zea mays</i> L.	<i>Poaceae</i>		x	MY-JU	h	PL	x	x	x
<i>Ziziphus jujuba</i> Mill.	<i>Rhamnaceae</i>	x	x	M-MY	st	PL	x	x	x
<i>Ziziphus lotus</i> (L.) Lam.	<i>Rhamnaceae</i>	x	x	MY-JU	sh	L	x		
Total							235	252	211

N: Nectar, P: Pollen, FP: Flowering period, JA: January, F: February, M: March, A: April, MY: May, J: June, JU: July, O: August, S: September, OC: October, N: November, D: December; MT: Morphological type, st: small tree, h: herb, t: tree, l: liana, sh: shrub, ssh: sub-shrub; C: Category, L: Local, PL: Planted. St: Station

2.2. Classes and families of inventoried honey plants

The distribution by class of the plants listed indicates that a very large number of species belong to the Dicotyledons, 270 species against 24 species belonging to Monocotyledons and 2 species to Pinopsida (Table 7).

Table 7. Classes of melliferous plants.

Classes	Number of families	Number of species	Percentage (%)
Dicotyledon	56	270	91.22
Monocotyledon	11	24	8.11
Pinopsida	2	2	0.67

The inventory carried out in the J ijel region made it possible to count 296 honey-producing plants belonging to 69 families (Table 8). The systematic spectrum shows the dominance of the Asteraceae family with 42 (14.2%) species, followed by the Fabaceae, Apiaceae, Rosaceae and Lamiaceae families with 40 (13.5%), 16 (5.4%), 15 (5%) and 14 (4.7%) species, respectively, while the rest of the families are less represented ranging from 1 to 10 species. Likewise, the most abundant genera are *Trifolium* (8), *Ranunculus* (8), *Silene* (7), *Solanum* (7), *Prunus* (6), *Convolvulus* (5), *Lotus* (5), *Iris* (4), *Lavatera* (4) and *Medicago* (4). Differently, Sekine et al., 2013 stated 66 families of melliferous plants dominated by Asteraceae (9.48%), Myrtaceae (7.11%), Solanaceae (5.69%), Malpighiaceae (4.27%), Bignoniaceae (5.21%) and Fabaceae (3.79%) and *Solanum* (seven species) and *Eucalyptus* (four species) were the dominant genera. Toopchi-khosroshahi and Lotfalizadeh, 2011 reported 22 families dominated by Fabaceae 16 species (16.32%), Asteraceae 14 species (14.28%), Lamiaceae 8 species (8.16%), Rosaceae 7 species (7.14%), Apiaceae 7 species (7.14%), Brassicaceae 6 species (6.12%), Papaveraceae 6 species (6.12%), Scrophulariaceae 6 species (6.12%). Nguemo et al., 2004 found 33 families of the identified melliferous plants dominated by Asteraceae (12.9%), Solanaceae (8.6%), Euphorbiaceae (7.6%), Malvaceae (6.4%), Myrtaceae (6.4%), Mimosaceae (5.1%) and Fabaceae (5.1%). Taha, 2015 has been able to report 24 families of melliferous plants dominated by Asteraceae (12), Fabaceae (10), Brassicaceae (9), Cucurbitaceae (8), Rutaceae (7). According to the results from different regions, the melliferous plants belong to Asteraceae family are worldwide spread melliferous plants. The palynological studies also indicated the importance of Asteraceae family to different bee species (Novais and Navarro, 2012).

Table 8. Families' diversity of melliferous plants.

Family	Number of species	Percentage (%)	Class
Acanthaceae	1	0.34%	Magnoliopsida
Aizoaceae	1	0.34%	Magnoliopsida
Alismaceae	1	0.34%	Magnoliopsida
Amaryllidaceae	2	0.68%	Liliopsida
Anacardiaceae	2	0.68%	Magnoliopsida
Apiaceae	16	5.41%	Magnoliopsida
Apocynaceae	2	0.68%	Magnoliopsida
Arecaceae	1	0.34%	Liliopsida
Asparagaceae	1	0.34%	Liliopsida
Asteraceae	42	14.19%	Magnoliopsida
Betulaceae	1	0.34%	Magnoliopsida
Bignoniaceae	1	0.34%	Magnoliopsida
Boraginaceae	6	2.03%	Magnoliopsida
Brassicaceae	6	2.03%	Magnoliopsida
Cactaceae	1	0.34%	Magnoliopsida
Campanulaceae	2	0.68%	Magnoliopsida
Caprifoliaceae	2	0.68%	Magnoliopsida
Caryophyllaceae	8	2.70%	Magnoliopsida
Cistaceae	2	0.68%	Magnoliopsida
Convolvulaceae	6	2.03%	Magnoliopsida
Cucurbitaceae	6	2.03%	Magnoliopsida
Cupressaceae	1	0.34%	Pinopsida
Dioscoreaceae	1	0.34%	Liliopsida
Dipsacaceae	2	0.68%	Magnoliopsida
Ericaceae	2	0.68%	Magnoliopsida
Euphorbiaceae	3	1.01%	Magnoliopsida
Fabaceae	38	12.84%	Magnoliopsida
Fagaceae	2	0.68%	Magnoliopsida
Gentianaceae	2	0.68%	Magnoliopsida
Geraniaceae	7	2.36%	Magnoliopsida
Hyacinthaceae	1	0.34%	Liliopsida
Hypericaceae	1	0.34%	Magnoliopsida
Iridaceae	6	2.03%	Liliopsida
Juglandaceae	1	0.34%	Magnoliopsida
Lamiaceae	14	4.73%	Magnoliopsida
Liliaceae	8	2.70%	Liliopsida
Linaceae	1	0.34%	Magnoliopsida
Lythraceae	3	1.01%	Magnoliopsida
Malvaceae	7	2.36%	Magnoliopsida
Meliaceae	1	0.34%	Magnoliopsida
Musaceae	1	0.34%	Liliopsida
Myrtaceae	2	0.68%	Magnoliopsida
Nyctaginaceae	2	0.68%	Magnoliopsida
Oleaceae	3	1.01%	Magnoliopsida
Onagraceae	1	0.34%	Magnoliopsida
Oxalidaceae	2	0.68%	Magnoliopsida
Papaveraceae	5	1.69%	Magnoliopsida
Pinaceae	1	0.34%	Pinopsida
Poaceae	1	0.34%	Liliopsida
Portulacaceae	1	0.34%	Magnoliopsida

Family	Number of species	Percentage (%)	Class
Punicaceae	1	0.34%	Magnoliopsida
Ranunculaceae	9	3.04%	Magnoliopsida
Resedaceae	1	0.34%	Magnoliopsida
Rhamnaceae	2	0.68%	Magnoliopsida
Rosaceae	15	5.07%	Magnoliopsida
Rubiaceae	2	0.68%	Magnoliopsida
Rutaceae	4	1.35%	Magnoliopsida
Salicaceae	2	0.68%	Magnoliopsida
Scrophulariaceae	6	2.03%	Magnoliopsida
Smilacaceae	1	0.34%	Liliopsida
Solanaceae	10	3.38%	Magnoliopsida
Tamaricaceae	1	0.34%	Magnoliopsida
Thymelaeaceae	1	0.34%	Magnoliopsida
Tiliaceae	1	0.34%	Magnoliopsida
Ulmaceae	2	0.68%	Magnoliopsida
Valerianaceae	1	0.34%	Magnoliopsida
Verbenaceae	4	1.35%	Magnoliopsida
Vitaceae	2	0.68%	Magnoliopsida
Xanthorrhoeaceae	1	0.34%	Liliopsida
69	296	100	3

2.3. Morphological types

The herbs dominating the morphological types of inventoried melliferous plants in the region of Jijel by 208 species (70%), followed by shrubs 28 species (10%), trees 21 species (7%), small trees and liana 16 species (5.5%) and sub-shrub 7 species (2%) (Figure 3). Nguemo et al., 2004 found. 36,5% herbs, 25,9% trees, 20,7% small trees and 16,9% shrubs. Taha et al., 2017 reported 76 (69.10%) were herbs, 29 (26.36%) trees, and 5 (4.54%) shrubs. Otherwise, Sekine et al., 2013 found 53 trees (25.5%), 41 herbs (19.7%), 40 shrubs (19.2%), 31 climbers (15.0%), 12 sub-shrubs (5.8%) and 12 small trees (5.8%). Delphine et al., 2017 reported trees (70.59%) and shrubs (29.41%). Adgaba et al., 2017 reported 61% shrubs, 27.67% herbs and 11.53% trees of melliferous plants of Al-Baha region in Saudi Arabia. Thus, the dominant botanical types vary from a region to another. The microclimatic environments formed by Phanerophytes have a big influence on the physiology of herbs and shrubs in that region. In addition, it is easy for wild herbs to survive and develop in the areas that are subjected to intermittent flooding and anthropogenic action (Mačukanović-Jocić and Jarić, 2016). In this study, the inventoried regions were apparently subjected to anthropogenic action such as agriculture and/or urbanization, which made them more suitable for herbs to install and grow compared to others botanical types.

On the other hand, the distribution of morphological types of melliferous flora by stations are represented in Figure 6 and are as follows:

- The 6m zone contained 167 (71%) herb species, 23 (10%) shrub species, 14 (6%) tree and liana species, 10 (4%) small tree species and 7 (3%) sub-shrub species.
- The 70m zone had 175 (69.5%) herb species, 25 (10%) shrub species, 17 (6.5%) tree species, 15 (6%) small tree species, 14 (5.5%) liana species and 6 (2.5%) sub-shrub species.
- The 700m zone contained 146 (69%) herb species, 22 (10.5%) shrub species, 13 (6%) tree and liana species, 11 (5.5%) small tree species and 6 (3%) sub-shrub species.

According to these results, the percentage of melliferous herbs, shrubs, trees, lianas and sub-shrubs were about the same in all stations. On the other hand, the small trees had less percentage in the 6m zone (4%) than 70m and 700m zones, (6%) and (5.5%), respectively.

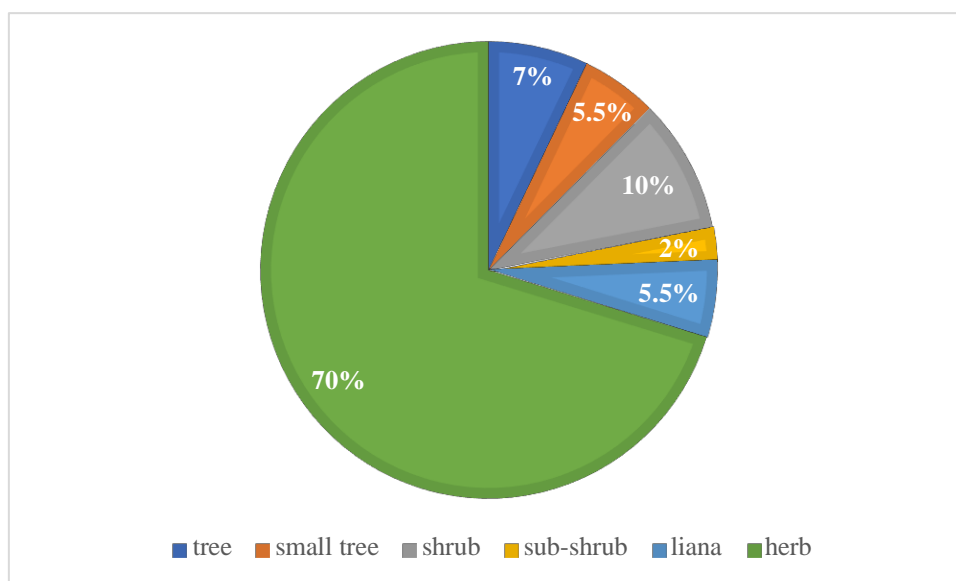


Figure 6. Morphological types of melliferous flora of Jijel. 5.5%: small tree, 70%: herb, 7%: tree, 5.5%: liana, 10%: shrub, 2%: sub-shrub

With 135 over 208 (65%) herb species were found in the three zones, the herbs were the most abundant morphological type between the three stations. However, small trees are the most changing morphological types between the different stations, with 8 over 16 (50%) small trees species were found in the three zones. In addition, 16 over 28 (57%) shrub species and 12 over 21 (57%) tree species were found in the three zones. On the other hand, 6 over 7 (85%) sub-shrubs,

Solanum sodomaeum L. was only found at 6m zone, are found in all zones. In addition, 12 over 16 (75%) inventoried liana were found in the three zones, while *Citrullus lanatus* (Thunb.) Matsum. & Nakai and *Cucumis melo* L. were only found in 6m zone, *Ipomoea tricolor* Cav. was found only in 70m zone and *Solanum dulcamara* L. was only found in 70 m and 700m zones.

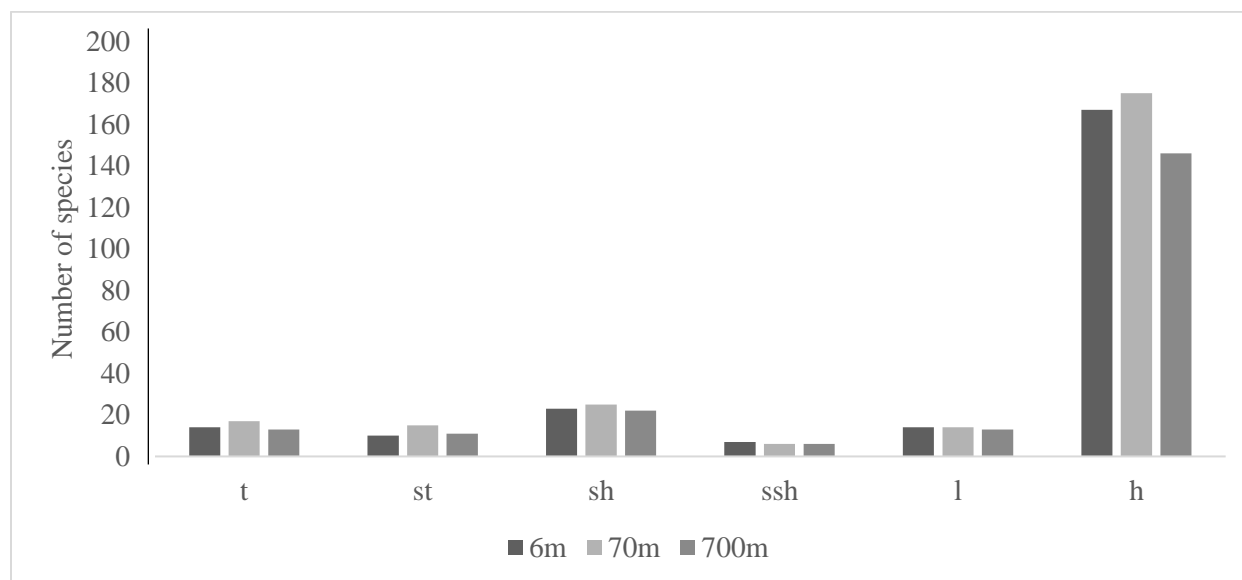


Figure 7. Distribution of morphological types according to the stations. st: small tree, h: herb, t: tree, l: liana, sh: shrub, ssh: sub-shrub: 6m: 6m zone, 70m: 70m zone, 700m: 700m zone, FT: Total flora.

2.4. Planted flora

Generally, the apiaries of Jijel region are close to the anthropized zones, which more likely make presence of planted plants. The number of planted melliferous plants inventoried in this region is 68 species (23%) (Table 1). The 70m zone contains more species than the other zones by 64 species then the coastal zone by 52 species and finally the montane zone by 50 species. Sekine et al., 2013 identified 34 species (16.1%) exotic melliferous species. Bhalchandra et al., 2014 reported 29 (55.8%) melliferous plants as agricultural in Nasik district (M. S.) India. Nguemo et al., 2004 found (64%) of the total melliferous plants as planted in Western Cameroon.

Jijel region has agricultural and rural areas that participate by 23% of the region's honey flora. In addition, the proximity of apiaries to houses leads to the presence of horticultural and ornamental species. The number of plants found in all stations is 47 (70%), which means that these species are the most planted in almost all Jijel region. This high percentage of similarity between planted flora is because they

are subjective to the human action and choice, so their distribution is changeable and irregular. The 70m station has larger number than other stations due to their proximity to the village signifying the increase of the anthropic action in this zone.

Within the 68 planted melliferous plants, herbs were 24 (35%) species, small trees were 16 (23.5%) species, trees were 12 (17.5%) species, liana were 8 (12%) species, shrubs were 6 (9%) species and sub-shrubs were 2 (3%) species (Table 9). The herbs are still the most abundant morphological type, however, they just represented 35% of the total planted flora. In addition, small trees were the second most abundant morphological type by 16 species, which means that all small trees that were inventoried were planted plants.

Table 9. Distribution of morphological types of planted melliferous flora.

	Herb	Tree	Small tree	Shrub	Sub-shrub	Liana
Number	24	12	16	6	2	8
Commun species	21	6	9	4	2	5

2.5. Flowering schedule

It is necessary for the flowering timing of plants to be in harmony with environmental conditions for the sake of prosperous reproduction (Jung und Müller, 2009). The flowering time influences pollination, determine the timing of seed ripening and dispersal and affect the animals that rely on pollen, nectar and seed as the prime food sources (Fitter and Fitter, 2002). The process that prevent plants from flowering in the unfavorable period of winter cold is known as vernalization (Kim at al., 2009). Flowering rhythms are particularly important; they condition the growth of the colony, the swarming and the constitution of the winter reserves. The discrepancies between the rhythm of flowering and the biological rhythm of the colony are enough to create an imbalance often resulting in the disappearance of the colony (Maurizio and Louveaux, 1960). In Jijel region, melliferous plants flowering occurs the whole year with a variation in the number of the species flowered during the various months. We found that the most of honey plants bloom in May with 236 species, then April with 212 species, followed by June with 168 species, July 104, March 103, August 95, September 71, October 60, November 55, February 42 and lastly January by 37 species (Figure 5). Likewise, Taha et al., 2017 reported that March had 64 blooming species more than any other month and January had the lowest numbers of blooming species (7). Albaba, 2015 noticed that in

April more melliferous plants are blooming (111), followed by May (102), March (76) June (73) and lastly December (9).

The daylength triggers the initiation of flowering in many plant species (Samach and Coupland, 2000). Long-day (LD) plants are the plants that flower during the lengthening days (spring or early summer) and short-day (SD) are the plants that flower during the shortening days (late summer or autumn) (Kim et al., 2009). The flowering periods of most inventoried honey plants spread between April and June and the lowest flowering period of honey plants is recorded between December and February, which means the dominance of (LD) plants. Therefore, the bee colonies take maximum advantage in April, May and June to increase the colony size and to produce the honey and store it to resist the period of scarcity between December and February. Bista and Shivakoti, 2001 reported that Mid-Nov - Feb (winter season) and June - Aug (rainy season) were identified as the dearth periods for honeybee at Kabre area in Nepal. Antonie, 2014 reported that the most favorable period for honeybees to forage is from May to June in Sebeş (Nepal). The blooming period of a plant can change according to the type of soil, climatic factors and the habitat of the vegetation (Rodinov and Shabanshov, 1986). In temperate climates, plants enter in the vernalization process during cold winter to ensure flowering in favorable season (Jung und Müller, 2009).

The category of plants that bloom for three months is predominant by 119 species then for 2 months by 53 species, for 4 months by 48 species and the other categories by less than 20 species each one while there are no species blooms only for a month (Figure 6). Among the plants that flower during the four seasons, we found *Bougainvillea glabra* Choisy, *Citrus limon* (L.) Burm.f., *Lobularia maritima* (L.) Desv., *Rosmarinus officinalis* L., *Musa* sp., *Pelargonium* sp. and *Solanum nigrum* L. Among the plants that bloom for three seasons, there are *Andryala integrifolia* L., *Daucus carota* L., *Lantana camara* L., *Lippia citriodora* Kunth, *Melia azedarach* L., *Ricinus communis* L., *Verbena officinalis* L. Examples of plants that bloom for two seasons are *Bellis annua* L., *Cestrum nocturnum* L., *Picris echioides* L., *Echium vulgare* L., *Eucalyptus* sp. *Mentha pulegium* L. Plants that bloom during a season such as *Acacia cyanophylla* Lindl., *Acanthus molis* L., *Allium triquetrum* L., *Asphodelus microcarpus* Viv. *Carlina racemosa* L., *Crataegus monogyna* Jacq., *Salvia verbenaca* (L.) Lighter. In addition, there are two species with two flowering periods during the year (*Capsella bursa-pastoris* (L.) Medik and (*Phaseolus vulgaris* L.). Nguemo et al., 2004 reported (14.1%) of inventoried melliferous plants bloom all the year. Therefore, the

presence of 40% of the species that flower for three months indicates that the blooming stage of the honey flora is changing during the year and indicates the change in foraging resources for bees during the year.

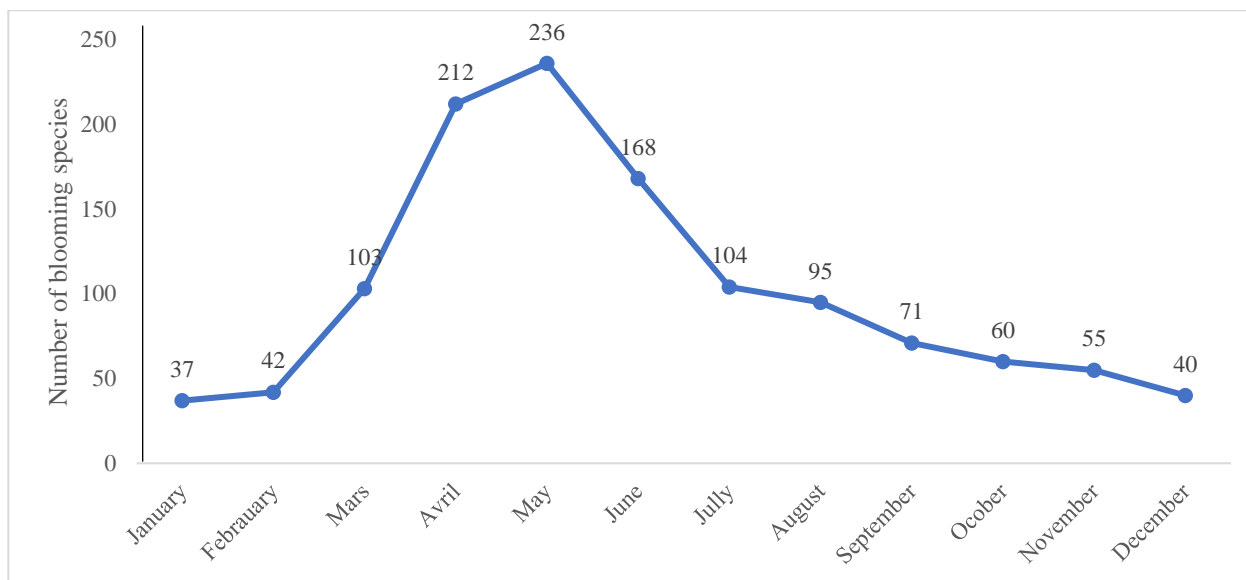


Figure 8. Number of blooming species during each month in Jijel region (2015-2017).

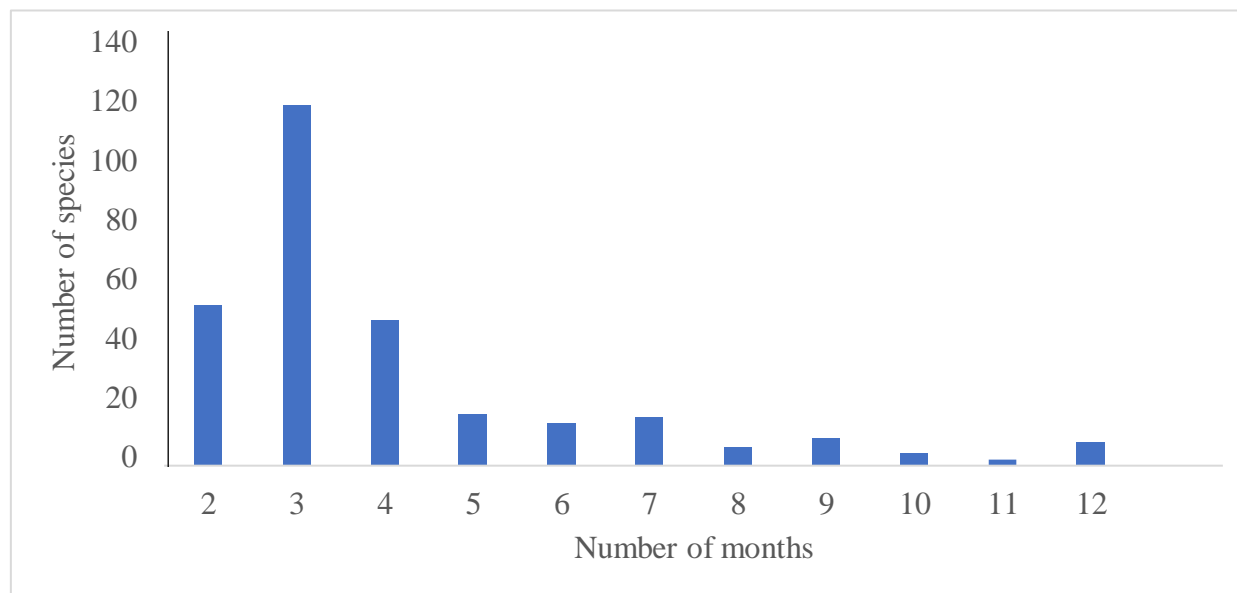


Figure 9. Duration of flowering time of melliferous plants in Jijel region (2015-2017).

Many plants species have flowers that are open permanently, however, other species, have flowers with alternated periods of opening and closure. These alternated periods happen due to a closure movement, petal withering or abscission (Doorn and Meeteren, 2003). The circadian rhythm of many flowers by opening during the day and closing at night is known as Linné's floral clock, which gives plants opportunity

for pollination according to the environmental conditions (Ke et al., 2018). The circadian clock of the flower of the melliferous flora in Jijel region were diverse. For example, the flowers of *Bellis sylvestris* Cyr. and *Ipomoea tricolor* Cav. open in the morning and close in the afternoon, *Mirabilis jalapa* L., *Cestrum nocturnum* L. and *Iris sisyrinchium* L. open in the afternoon, *Oxalis corniculata* L. open at 10 a.m. and close just after noon, the *Vinca difformis* Pourr. and *Rubus ulmifolius* Schott are always open. The flowering spectrum of the honey flora in this region is also variable, depending on climatic conditions, for instance, the flowers of *Oxalis corniculata* L. do not open if the weather is cloudy. Humidity, light and temperature are the most important environmental cues that affect the opening and the closure of flowers (Doorn and Kamdee, 2014).

2.6. Beekeeping value of honey flora

Nectar and pollen are the main food sources for bees to survive and develop hives. Nectar is the most important raw material used by bees to make honey (Pain and Mangenet, 1966). The secretion of nectar and its quality vary with the age of the flowers and during the day. The sugar content of linden nectar (*Tilia cordata*), for example, drops during flowering from 42% to 26%. Linden nectar is primarily secreted in the evening and at night, while in wild chicory (*Cichorium intybus*), it is produced only in the first half of the day (Fluri et al., 2001). The amount of nectar varies greatly depending on climate, soil and altitude. For a given variety the quantity of nectar secreted is the result of the degree of mineral absorption by plants and its photosynthetic activity (Philippe, 1998). On the other hand, the pollen is also the main food source for honeybee brood from larvae to young adults. The more flowers are abundant, the more the honeybees select the pollens that provide them with the maximum protein (Maurizio, 1953). Melliferous flora of the study area contains 206 plants that are both nectariferous and polliniferous, 49 polliniferous plants and 41 nectariferous plants (Figure 7). Plants that produce nectar and pollen represent 70% of the total honey flora while the number of polliniferous plants is greater than nectariferous plants by 8 species. Likewise, all stations follow the same trend to have plants that are both nectariferous and polliniferous dominate the scene (Figure 7). Therefore, the dominance of plants that are both nectariferous and polliniferous helps honeybees to have better food resources and minimize the amount of time and energy used to travel between flowers foraging nectar and pollen. In the West Bank Governorates, Palestine, the inventoried melliferous plants by Albaba, 2015 were 20% of as nectar, 12% as pollen and 68% as nectar and pollen yielding plants. Toopchi-khosroshahi and

Lotfalizadeh, 2011 reported 21 species (21.42%) were as nectar source, 33 species (33.67%) as pollen source and 44 species (44.89 %) as nectar and pollen source. Otherwise, the inventoried melliferous plants in Northwest of Benin by Ahouandjinou et al., 2017 were 60.5% of as nectar source, 12.8% as pollen source and 23.3% as nectar and pollen source. Nguemo et al., 2004 were able to find higher percentage of pollen yielding plants with 41%, then 23% nectar sources and then 16% as nectar and pollen source.

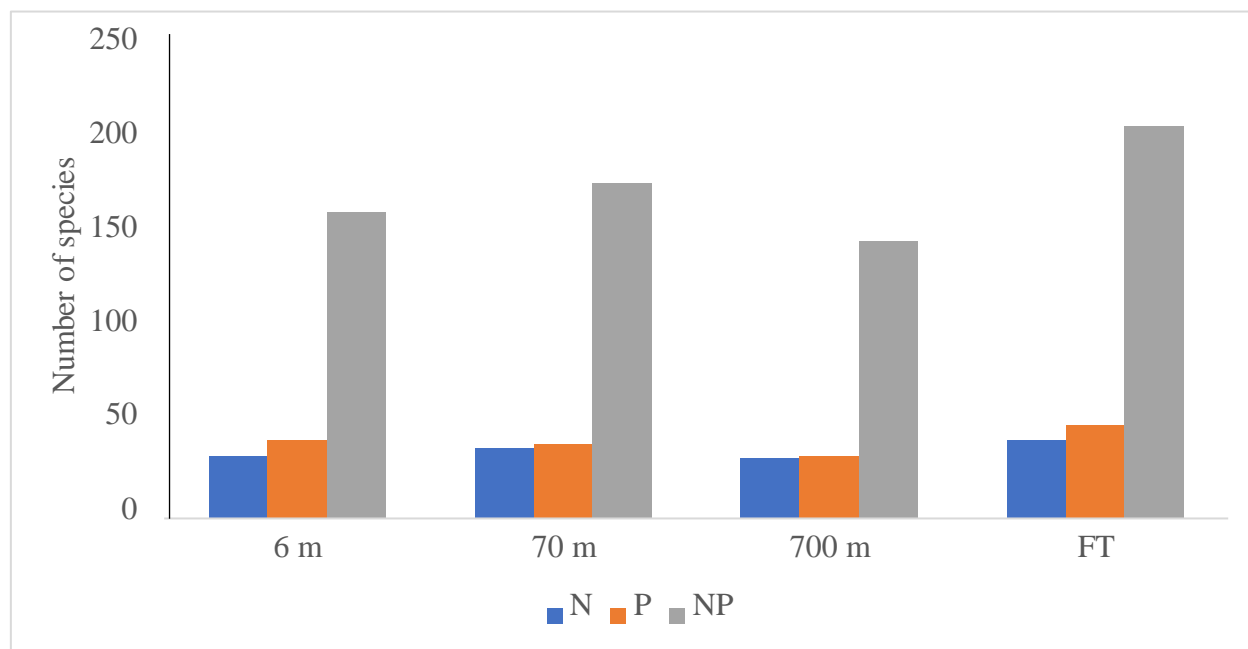


Figure 10. Distrubution of polliniferous and nectariferous plants in the region of Jijel. 6 m: 6m zone, 70 m: 70m zone, 700 m: 700m zone, FT: Total flora; N: Nectariferous, P: Polliniferous, NP: Nectariferous and polliniferous.

2.7. Abundant and nearby honey plants

According to Pesson and Louveau (1984), the closest and the most abundant food sources are exploited preferentially by honeybees. Therefore, it is important to identify the melliferous plants that have the most effect on the honey produced in each station.

2.7.1 Abundant and nearby melliferous plants of 6m zone

The station at 6 m above sea level contains 21 most important melliferous plants (Table 10). Among these plants, the wild flora is represented by 16 (75%) species and the planted flora is represented by 5 species (25%). These plants are represented by 2 nectariferous plants, 2

polliniferous plants and 17 nectariferous and polliniferous plants. The presence of 21 large-foraging melliferous plants in this region among them there are 19 plants produce nectar and 19 plants produce pollen means that these plants have a significant influence on honey characteristics produced in this region and high probability for poly floral honey. In addition, we can obtain a honeydew honey by the presence *Eucalyptus* and *Populus nigra* which are good sources of honeydew. The anthropogenic effect is represented by five species (*Citrullus lanatus*, *Cucurbita pepo*, *Eucalyptus*, *Fragaria × ananassa*, *Opuntia ficus-indica*). Among these species, two species (*Eucalyptus* and *Opuntia ficus-indica*) are potentially present each year but the other species can be replaced by other species since they are planted each year. In this zone, May, April and June, respectively have 18, 16 and 15 species in bloom, are the most important months for bees to find food because most of the honey plants flourish in these months. These melliferous plants bloom throughout the year. However, the months of January and December are times of scarcity for bees since the plants that bloom in these months (*Fragaria × ananassa* and *Lobularia maritima*) are not good sources of nectar.

Table 10. Abundant and nearby melliferous plants of 6m zone.

Melliferous plants	Flowering	Apiarian benefits	70m	700m
<i>Citrullus lanatus</i> (Thunb.) Matsum. & Nakai	My-O	nn,pp		
<i>Cucurbita pepo</i> L.	A-N	N, P	x	x
<i>Daucus carota</i> L.	M-N	n-p	x	x
<i>Echinops spinosus</i> L.	My-JU	N, p		
<i>Eryngium maritimum</i> L.	J-S	N, p		
<i>Eucalyptus</i> sp.	M-O	nn, p, H	x	x
<i>Fragaria × ananassa</i> (Weston) Duchesne ex Rozier	JA-My	n, p	x	x
<i>Galactites tomentosa</i> Moench	M-JU	nn, p	x	x
<i>Hedypnois cretica</i> (L.) Dum.Cours.	A-My	n,p		
<i>Hedysarum coronarium</i> L.	A-J	N, P	x	x
<i>Iris foetidissima</i> L.	M-J	N		
<i>Lobularia maritime</i> (L.) Desv.	S-My	P	x	x
<i>Lotus cytisoides</i> L.	F-J	N, p	x	x
<i>Ononis variegata</i> L.	A-J	N		
<i>Opuntia ficus-indica</i> (L.) Mill.	A-J	nn,pp	x	x
<i>Phyla nodiflora</i> (L.) Greene	M-O	N		
<i>Populus nigra</i> L.	F-A	p, Pr, H	x	x
<i>Retama monosperma</i> (L.) Boiss.	M-My	n, p		
<i>Rubus ulmifolius</i> Schott	A-N	nn, pp	x	x
<i>Ziziphus lotus</i> (L.) Lam.	M-JU	N, p		

N: High nectar, nn: Medium nectar, n: Low nectar: P: High pollen, pp: Medium pollen, p: Low pollen, H: Honeydew, Pr: Propolis, JA: January, F: February, M: March, A: April, MY: May, J: June, JU: July, O: August, S: September, N: November.

2.7.2. Abundant and nearby melliferous plants of 70m zone

The station at 70 m above sea level contains 22 most important melliferous plants (Table 11). Among these plants, spontaneous flora is represented by 15 (68%) species and the planted flora is represented by 7 (32%) species. These plants are represented by 4 polliniferous plants and 18 nectariferous and polliniferous plants. The presence of 22 melliferous plants with large-foraging sources in this region among them there are 18 plants produce nectar and 22 plants produce pollen means that these plants have a significant influence on the characteristics of honey produced in this region and high probability for poly floral honey. In addition, we can obtain honeydew honey by the presence of the trees that produce honeydew (*Eucalyptus*, *Prunus domestica* and *Quercus suber*). The anthropogenic effect is represented by seven species (*Citrus aurantium*, *Citrus sinensis*, *Eucalyptus*, *Lantana camara*, *Olea sp*, *Prunus domestica* and *Robinia pseudacacia*). In this area,

Table 11. Abundant and nearby melliferous plants of 70m zone.

Melliferous plants	Flowering	Apiarian benefits	6m zone	700m zone
<i>Bellis annua</i> L.	D-My	P	x	x
<i>Calicotome spinosa</i> (L.) Link.	M-My	n,p	x	x
<i>Citrus aurantium</i> L.	A-My	N, pp		
<i>Citrus sinensis</i> (L.) Osbeck	A-My	N, pp	x	x
<i>Crataegus monogyna</i> Jacq.	M-My	nn, p	x	x
<i>Erica arborea</i> L.	M-My	N, P	x	x
<i>Eucalyptus</i> sp.	M-O	nn, p, H	x	x
<i>Galactites tomentosa</i> Moench	M-JU	nn, p	x	x
<i>Hedysarum coronarium</i> L.	A-J	N, P	x	x
<i>Inula viscosa</i> (L.) Aiton	S-N	nn, pp	x	x
<i>Lantana camara</i> L.	A-D	Nn	x	x
<i>Lotus cytisoïdes</i> L.	F-J	N, p	x	x
<i>Mentha pulegium</i> L.	My-S	N, p	x	x
<i>Myrtus communis</i> L.	A-My	nn,pp	x	x
<i>Olea</i> sp.	M-J	P	x	x
<i>Oxalis pes-caprae</i> L.	D-M	N, p	x	x
<i>Pistacia lentiscus</i> L.	F-M	P	x	x
<i>Prunus domestica</i> L.	M-A	N, P, H	x	x
<i>Quercus suber</i> L.	A-My	p, H	x	x
<i>Robinia pseudacacia</i> L.	A-J	N, p		
<i>Rubus ulmifolius</i> Schott	A-N	nn, pp	x	x
<i>Scilla maritima</i> L.	O-Oc	nn.p	x	x

N: High nectar, nn: Medium nectar, n: Low nectar: P: High pollen, pp: Medium pollen, p: Low pollen, H: Honeydew; JA: January, F: February, M: March, A: April, MY: May, J: June, JU: July, O: August, S: September, OC: October, N: November, D: December.

honey plants bloom throughout the year and there is no real time of scarcity for bees. April, May and March respectively have 17, 17 and 11 species in bloom, are the most important months for bees to stock up as most of the honey plants flourish in these months.

2.7.3. Abundant and nearby melliferous plants of 700m zone

The station at 700 m above sea level contains 25 most important melliferous plants (Table 12). Spontaneous flora is represented by 17 (68%) species and the planted flora is represented by 8 (32%) species. These plants are represented by 1 nectariferous plant, 4 polliniferous plants and 20 nectariferous and polliniferous plants. The presence of 25 large-foraging melliferous plants in this region among them there are 21 plants produce nectar and 24 plants produce pollen means that these plants have a significant influence on characteristics of honey produced in this region and high probability for poly floral honey. In addition, we can obtain honeydew honey by the presence of *Castanea sativa*, *Malus pumila*, *Prunus avium*, *Prunus domestica*, *Pyrus communis* and *Quercus suber*, which are good sources of honeydew. The anthropogenic effect is represented by eight species of fruit trees (*Malus pumila*, *Olea sp*, *Prunus avium*, *Prunus domestica*, *Prunus dulcis*, *Prunus persica*, *Punica granatum* and *Pyrus communis*). These 25 plants bloom for 11 months and there is no flowering plant in January. April, March and May respectively have 20, 15 and 14 species in bloom, are the most important months for bees to stock up as most of these honey plants flourish in these months. However, the months of January and December are times of scarcity for honeybees since there is no flowering plant in January and the presence of a single plant (*Daphne gnidium*) which is at the end of flowering in December.

Table 12. Abundant and nearby melliferous plants of 700m zone.

Melliferous plants	Flowering	Apiarian benefits	6m zone	70m zone
<i>Allium triquetrum</i> L.	M-A	n.p	x	x
<i>Calicotome spinosa</i> (L.) Link.	M-My	n,p	x	x
<i>Castanea sativa</i> Mill.	A-J	nn, pp, H		
<i>Cistus salviaefolius</i> L.	A-My	P	x	x
<i>Cytisus triflorus</i> Lam.	F-A	Nn	x	x
<i>Daphne gnidium</i> L.	JU-D	nn, p	x	x
<i>Erica arborea</i> L.	M-My	N, P		
<i>Galactites tomentosa</i> Moench	M-JU	nn, p	x	x
<i>Genista ulicina</i> Spach	A-My	n,p		x

Melliferous plants	Flowering	Apiarian benefits	6m zone	70m zone
<i>Inula viscosa</i> (L.) Aiton	S-N	nn, pp	x	x
<i>Lavandula stoechas</i> L.	F-My	N,p	x	x
<i>Lotus cytisoïdes</i> L.	F-J	N, p	x	x
<i>Malus pumila</i> Mill.	A-My	N, p, H	x	x
<i>Myrtus communis</i> L.	A-My	nn,pp	x	x
<i>Olea</i> sp.	M-J	P	x	x
<i>Pistacia lentiscus</i> L.	F-M	P	x	x
<i>Prunus avium</i> (L.) L.	M-A	nn, P, H ; Pr		x
<i>Prunus domestica</i> L.	M-A	N, P, H	x	x
<i>Prunus dulcis</i> (Mill.) D.A.Webb, 1967	F-A	N, pp		x
<i>Prunus persica</i> (L.) Batsch	M-A	Pp, M	x	x
<i>Punica granatum</i> L.	My-O	nn, p	x	x
<i>Pyrus communis</i> L.	M-A	nn, p, H	x	x
<i>Quercus suber</i> L.	A-My	p, H	x	x
<i>Rubus ulmifolius</i> Schott	A-N	nn, pp	x	x
<i>Trifolium campestre</i> Schreb.	A-J	N, pp	x	x

N: High nectar, nn: Medium nectar, n: Low nectar: P: High pollen, pp: Medium pollen, p: Low pollen, H: Honeydew, Pr: Propolis; JA: January, F: February, M: March, A: April, MY: May, J: June, JU: July, O: August, S: September, OC: October, N: November, D: December.

2.7.4. Similarities between the stations

From the lists of the most important honey plants in the three stations, we found that there are only three honey plants spread on a large scale in the different stations of this region (*Galactites tomentosa* Moench, *Lotus cytisoïdes* L. and *Rubus ulmifolius* Schott). However, there are 11 common species between 70m and 700m zones (*Calicotome spinosa* (L.) Link., *Erica arborea* L., *Galactites tomentosa* Moench, *Inula viscosa* (L.) Aiton, *Lotus cytisoïdes* L., *Myrtus communis* L., *Olea* sp., *Pistacia lentiscus* L., *Prunus domestica* L., *Quercus suber* L. and *Rubus ulmifolius* Schott). In addition, 6 species common between the coastal and 70m stations (*Eucalyptus* sp., *Galactites tomentosa* Moench, *Hedysarum coronarium* L., *Lotus cytisoïdes* L., *Oxalis pes-caprae* L. and *Rubus ulmifolius* Schott) and three species between the coast and mountain stations (*Galactites tomentosa* Moench, *Lotus cytisoïdes* L. and *Rubus ulmifolius* Schott). Therefore, it appears that at least three types of poly-floral honeys can be obtained in Jijel region. On the other hand, it is obvious that honeys characteristics of high altitudes are different compared to those of average altitudes and more different compared to those of low altitudes.

2.7.5. Flowering periods:

Flowering spectrum of these most important honey plants during the year is a little different in the three stations. Spring is the most suitable season for bees to obtain food and develop apiaries in the three stations with a preference in the high-altitude area that contains up to 20 flowering plants in April. Summer is the second favorite season for beekeeping especially in the low altitude area, which has more flowering plants . Winter is a season of provisions' lack for honeybees in the three stations and especially in the high-altitude station, which suffers a real period of scarcity and beekeepers feed honeybees colony with artificial food.

Chapter II.

Honey analyzes

1. Materials and methods

1.1. Samples

Twenty-two honey samples were collected from different regions in Jijel (Algeria). Half of these samples were collected from regions close to the Mediterranean Sea and the other half from mountain regions (Table 13). All these samples were produced from hives placed in areas with diverse vegetation in order to get poly-floral honeys that are more representable of this region. In addition, Honeys from hives placed in vast monoculture fields were avoided. All samples were stored at 4-5 °C in airtight glass containers until analyses.

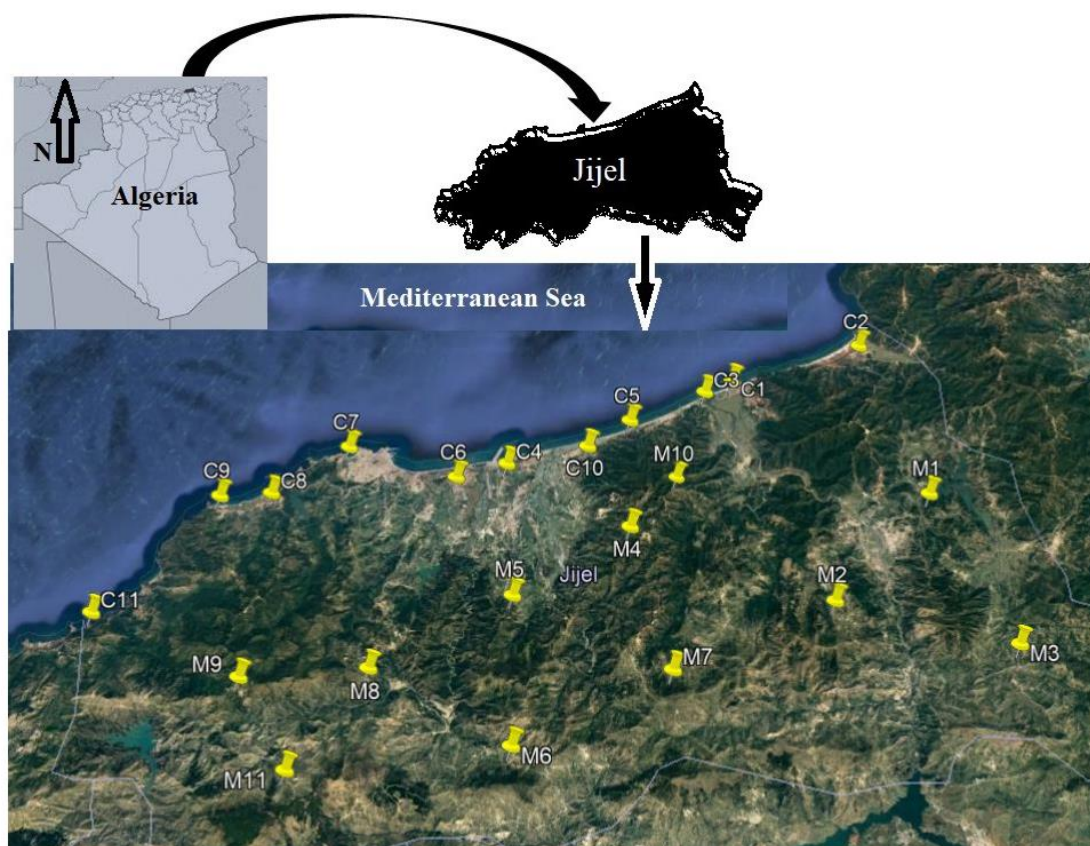


Figure 11. Geographical location of samples.

Table 13. Geographical information of samples.

Sample	Location	longitude	latitude	Altitude (m)
C1	Beni Belaid	6.115992	36.87588	6
C2	Oued Zhour	6.248999	36.90815	9

C3	El Janah	6.087643	36.86534	10
C4	Achouat	5.861033	36.80725	12
C5	El Balouta	6.010802	36.83923	30
C6	Boukhartoum	5.815903	36.79467	40
C7	Jijel	5.728306	36.81491	60
C8	El Kennar	5.655176	36.77435	60
C9	El Aouana	5.604307	36.7718	65
C10	Timizer	5.968151	36.81605	70
C11	Ziama Mansouriah	5.495001	36.67455	120
M1	El Milia	6.306215	36.77432	300
M2	Ouled Yahia	6.205439	36.68457	310
M3	Ghebala	6.377373	36.65082	330
M4	Bordj Thar	6.010342	36.74486	340
M5	Oudjana	5.897602	36.6885	400
M6	Djimla	5.900854	36.57211	510
M7	Ouled Askeur	6.049232	36.62948	520
M8	Taksana	5.765269	36.63066	570
M9	Selma	5.64602	36.62277	640
M10	Teyana	6.056851	36.78788	695
M11	Erraguen	5.697332	36.55349	700

C: Coastal and M: Mountainous; M: Meter.

1.2. Physicochemical analyses

The physicochemical analyses were determined according to the International Honey Commission (2009). In addition, the crystallization state of honey samples was observed.

1.2.1. Moisture content

Put homogenized honey sample in 50 ml flask. Afterward, close the flask and place it in water bath at 50 °C to dissolve the sugar crystals. After cooling, honey sample was stirred again for homogenization. And directly put the honey sample at a clean prism of a digital refractometer (Aqueous Lab) and read the measurement. The digital refractometer was already calibrated by distilled water at 20 °C at room temperature. The moisture content was expressed in g/100g.

1.2.2. pH

The pH of 10% honey solution was measured using calibrated pH-meter (HI 2210, Hanna).

1.2.3. Free, lactonic and total acidities

5 g of honey sample was dissolved in distilled water. Then, it was transferred to a 50 ml volumetric flask, added with distilled water to the mark and mixed. Afterward, 25 ml was pipetted in 250 ml beaker and initial pH (pHi) was measured. Stir gently with a bar magnet and titrate with the sodium hydroxide solution up to 10 ml, then with the sulphuric acid solution up to the second equivalence point.

$$\text{Free acidity (FA)} = V \times T \times (50/25) \times (1000/M)$$

T: the exact titre of the sodium hydroxide solution.

V: Note from the curve the free acidity neutralization volume in ml.

$$\text{The acidity of the lactones (LA)} = ((10-V) \times T - 0.05 \times V') \times (50/25) \times (1000/M)$$

V': the sodium hydroxide excess neutralization volume (corresponding to pH 7) in ml.

$$\text{The total acidity TA} = \text{FA} + \text{LA}$$

The FA, LA and TA were expressed in milliequivalents of sodium hydroxide required to neutralize 1 kg of honey.

1.2.4. Electrical conductivity (EC)

25 ml of 20% honey sample solution was poured into a beaker placed in thermostated water bath at 20 °C. After temperature equilibrium has been reached, the conductance was read in mS (HI 2315 Conductivity Meter, Hanna). Calculate the electrical conductivity of the honey solution, using the following formula:

$$\text{SH} = K.G$$

SH = electrical conductivity of the honey solution in mS.cm⁻¹

K = cell constant in cm⁻¹

G = conductance in mS

1.2.5. Ash

5 g of the sample was weighed to the nearest 0.001g into an ash dish (the ash dish was heated in the furnace at ashing temperature, afterward, was cooled in a desiccator to room

temperature and weighed to 0.001g). Add two drops of olive oil. Thereafter, water was evaporated using hotplate and commenced ashing without loss at a low heat rising to 400⁰ C. After the preliminary ashing, place the dish was put in the preheated furnace and heated for at least 1 hour. The ash dish was cooled in the desiccator and weighed. The ashing procedure was continued until constant weight is reached (m₁). The proportion of ash W_A in g/100g honey is calculated using the following formula:

$$W_A = 100 (m_1 - m_2) / m_0$$

m₀ = weight of honey taken

m₁ = weight of dish + ash

m₂ = weight of dish

1.2.6. Hydroxymethylfurfural (HMF)

A mass of 5 g of honey is dissolved in 25 ml of distilled water. 0.5 ml Carrez 1 solutions (15 g of potassium hexacyanoferrate (II), K₄Fe(CN)₆•3H₂O in 100 ml of distilled water) and Carrez 2 (30% zinc acetate solution) were added. The mixture was transferred to a 50 ml flask and the volume is adjusted with distilled water. After filtration, the first 10 ml of the filtrate are discarded, two 5 ml aliquots are then introduced into two test tubes, one with 5 ml of distilled water (analysis aliquot) and the other with 5 ml of 0.2% sodium bisulfite (reference aliquot). When the absorbance is greater than 0.6, the analysis and reference aliquots are diluted with distilled water and with the bisulfite sodium solution, respectively. Absorbance is read at 284 nm and 336 nm and the HMF content is given by the equation:

$$\text{HMF in mg/kg} = (A_{284} - A_{336}) \times 149.7 \times 5 \times D/W$$

A₂₈₄ = absorbance at 284 nm

A₃₃₆ = absorbance at 336 nm

D = Final volume of sample solution / 10.

W = mass in grams of the honey sample

1.3. Protein content

The protein content was analyzed according to the Bradford method reported by Azeredo *et al.* (2003). 0.1 ml solution of honey sample (50% w/v) was added 5 ml of Coomassie Brilliant Blue. The absorbance was measured at 595 nm (UV-1800 UV-Vis Spectrophotometer from

Shimadzu, Kyoto, Japan), against an albumin standard solution of bovine serum (0.1-1.4 mg/ml).

1.4. Color analysis

Color analysis was reviewed according to Ferreira *et al.* (2009). Honey in distilled water solutions of 50% (w/v) were centrifuged at 3000×g for 10 minutes centrifugation (centrifuge Model 3-16P, Sigma Laborzentrifugen GmbH, Osterode am Harz, Germany). The color was measured spectrophotometrically at 635 nm. The Pfund scale was used to classify the honeys as follows: $\text{mm Pfund} = -38.70 + 371.39 \times \text{Abs}$.

1.5. Total phenolic contents

The following method described by Bueno-Costa *et al.* (2016) was used to determine the total phenolic content (TPC): Honey solution of (0.1 g/mL) was centrifuged at 3000×g for 10 min. 0.5 mL of supernatant and 2.5 mL of 0.2 N Folin–Ciocalteu reagent were mixed for 5 min. Afterwards, 2 mL of sodium carbonate solution (75 g/L) was added and incubated for 2 h in dark. The absorbance was measured using a spectrophotometer at 765 nm. The TPC were expressed as mg gallic acid equivalent per 100 g of sample (mg GAE/100 g).

1.6. Total flavonoid contents

Total flavonoid content (TPC) was determined according method described by Kim *et al.* (2003) and Chaikham *et al.* (2016). Solution of honey in ddH₂O (1 mL; 0.5 g/mL) was mixed with 300 µL NaNO₂ (5.0%). A volume of 300 µL of AlCl₃ (10%) was added to the mixture, and after 6 min, 2 mL of 1M NaOH was added. A spectrophotometer was used at 510 nm to measure the absorbance. Standard curve was defined by known concentrations of quercetin (0-40 mg/l), and the results were expressed as mg quercetin equivalent per 100 g of sample (mg QE/100 g).

1.7. DPPH radical scavenging activity

Assay of 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity (RSA) was performed according to Meda *et al.* (2005) procedure. The 0.75 mL of each honey solution in methanol (2.5-160 mg/mL) was mixed with 1.5 mL of DPPH in methanol (0.02 mg/mL). After 15 minutes in the dark, the absorbance was measured at 517 nm. DPPH radical solution without sample was served as the blank sample. The results were calculated based on formula: %Inhibition = ((blank absorbance - sample absorbance)/blank absorbance) × 100. The half maximal inhibitory

concentration (IC₅₀) value of each honey sample was estimated from the plot of % inhibition vs. honey concentration.

1.8. Reducing power

The following method of reducing power (RP) determination was used (Küçük *et al.*, 2017): 1 mL of honey solution (5.0%) was added to 2.5 mL phosphate buffer (0.2 M, pH 6.6) and 2.5 mL 1% potassium ferric cyanide (K₃Fe (CN)₆). The mixture was incubated at 50 °C for 20 min. Afterwards, 2.5 mL 10% trichloroacetic acid was added, and the mixture was centrifuged at 3000×g for 10 min. The supernatant (2.5 mL) was mixed with 2.5 mL of distilled water and 0.5 mL of 0.1% FeCl₃. The absorbance was measured at 700 nm. Ascorbic acid (1.0 mg/mL) was used as a reference standard.

1.9. Antibacterial activity

Agar disc diffusion assay of 100%, 50% and 25% honey concentration (with distilled water) were used against three strains of bacteria, which were *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, and *Pseudomonas aeruginosa* ATCC 27853 (Pasteur Institute of Algeria, Algeria), according to Alderman and Smith (2001). The results were expressed in zone of growth inhibition (mm).

1.10. Pesticides

1.10.1. Pesticides analysis

The honey samples were dissolved in a 25 mL vial using Milli-Q water. After that, 3 mL of honey solution was introduced into 10 mL tube for the pesticides' extraction, 3 mL of ethyl acetate was added to the tubes. The tubes were stirred for 2 min and then centrifuged at 3000 rpm for 5 min. The upper organic phase was transferred to another 15 mL tube. The residues were extracted twice using the same method mentioned above. The entire organic phase was collected and dried with Na₂SO₄. The solvent was evaporated using a rotary evaporator under reduced pressure (40-50 torr), which lowers the boiling temperature of the constituents of the honey mixture and in particular the solvents (ethyl acetate 9.1 °C, water 34.0 °C). The residues were dissolved using 1 mL of ethyl acetate for the gas chromatography analysis.

The gas chromatograph (Shimadzu GC-14) was used for the determination of OPs

pesticides. It is equipped with a tritium electron capture detector (3H-ECD). A Megabore HP-608 polysiloxane column (30 m \times 0.53 mm id \times 0.5 μ m film thickness) was used. In the ECD detector, N₂ gas was used as make-up gas with a flow rate of 4 mL/min. The chromatographic separation was carried out in temperature programming mode. After the injection of the sample extract, the initial temperature of the oven (65 °C) was kept constant for 2 min and increased to 200 °C (30 °C/min), and then holding for 5 min. The oven temperature was finally increased again to 250 °C with an increment of 5 °C/min (hold for 15 min).

The limits of quantification (LOQs), varied from 0.05 to 1.3 LOQ (ng/g). The limit of detection (LOD) varied from 0.1–2.1 ng/g. For all the pesticides were in total agreement with the literature values reported for GC-ECD analysis of OPs pesticides (Tette et al., 2016; López et al., 2014; Mukherjee, 2009).

1.10.2. Photodegradation of organophosphorus (OPs) pesticides

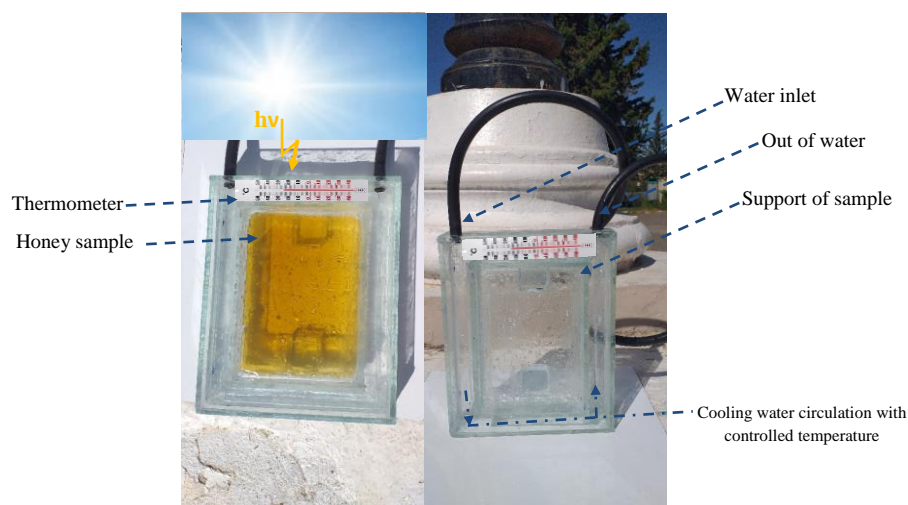


Figure 12. Experimental solar photoreactor support for honey photodegradation reaction.

The photodegradation experiments were conducted on a photocatalytic support exposed directly to sunlight (Figure 2). A CS320 digital thermopile pyranometer purchased from Campbell Scientific and Apogee Instruments was used to measure the global solar radiation. CS320 is suitable for applications for environmental research, agriculture and large mesoscale weather networks. The average irradiation flux measured during the experiments was 750 W/m² (Doufar

et al. 2020) while the reaction temperature reached 25 °C. The honey samples used in the photodegradation reaction were the three samples among the 22 listed in Table 17 that have high values for OPs pesticides; they were exposed to sunlight in June 2019; all experiments were performed in triplicate. The kinetics of photodegradation were studied during 1 h. Every 10 min, we collected a sample for analysis by GC-ECD.

1.11. Statistical analysis

All tests were performed in triplicate and the results were expressed as mean \pm standard deviation. The parameters of the descriptive statistics were calculated using the Microsoft Excel 2007 program. A one-way analysis of variance (ANOVA) was carried out with the STATISTICA 7.1 software to highlight the presence or absence of a significant difference between the samples of honey, which was considered statistically at the level of 0.05. The relationships between the parameters were determined by the correlation matrix ($p < 0.01$), while the means comparison between coast and mountain honeys were determined by Student's t-test using XLSTAT 2014.

2. Results and discussion

2.1. Physicochemical parameters of honeys

The crystallization of honey is more common on honeys that have relatively more glucose (Gleiter et al. 2006). Honey samples have F (fructose)/G (glucose) ratio greater than 1.33 stay liquid for a long time, however, the honey with a ratio less than 1.11 crystallizes quickly. In addition, other carbohydrates, moisture content, pollen, and air bubbles can act as seed crystals (Smanalieva and Senge 2009). On the other hand, Escuredo et al. (2014) have demonstrated that fructose, glucose, moisture content and sugar ratios (F + G, F/G and G/W) are the principle indicators of honey crystallization. Between the 22 tested samples, four samples were crystallized (M1, M4, M7 and M10) and the others were liquid. Al et al. (2009) reported three among 24 tested samples were crystallized. Crystallized samples were all mountainous, which could be due to the usage of sugar syrup by beekeepers during the dearth period in high altitude.

2.1.1. Moisture content

The moisture content is an important criterion to establish the quality of honey (Al et al. 2009). The moisture content of honey is related to different factors like the period of harvesting, ripening

process, and climatic conditions (Finola et al. 2007). During storage, higher water content could lead to undesirable honey fermentation (Ribeiro et al. 2014). The honey samples had a moisture content ranging from 16.7% to 19.8 % (Figure 1). The samples M3 (16.7) and M10 (16.7) had the lowest values and the sample C5 (19.8) had the higher value, which were acceptable values (under 20%) as defined by the Council of the European Union (2002). The results reported for the samples differed significantly ($p < 0.05$). These results showed that these honeys were ripe. Manzanares et al. (2017) found normal moisture values in all honey samples studied. Can et al. (2015) and Ouchemoukh et al. (2007) reported different moisture contents of 16-20% and 14.64-19.04%, respectively. The moisture content did not present a significant correlation with the altitude and the moisture content of coast samples was not significantly higher than mountain samples with 18.23 % against 17.8 %, respectively.

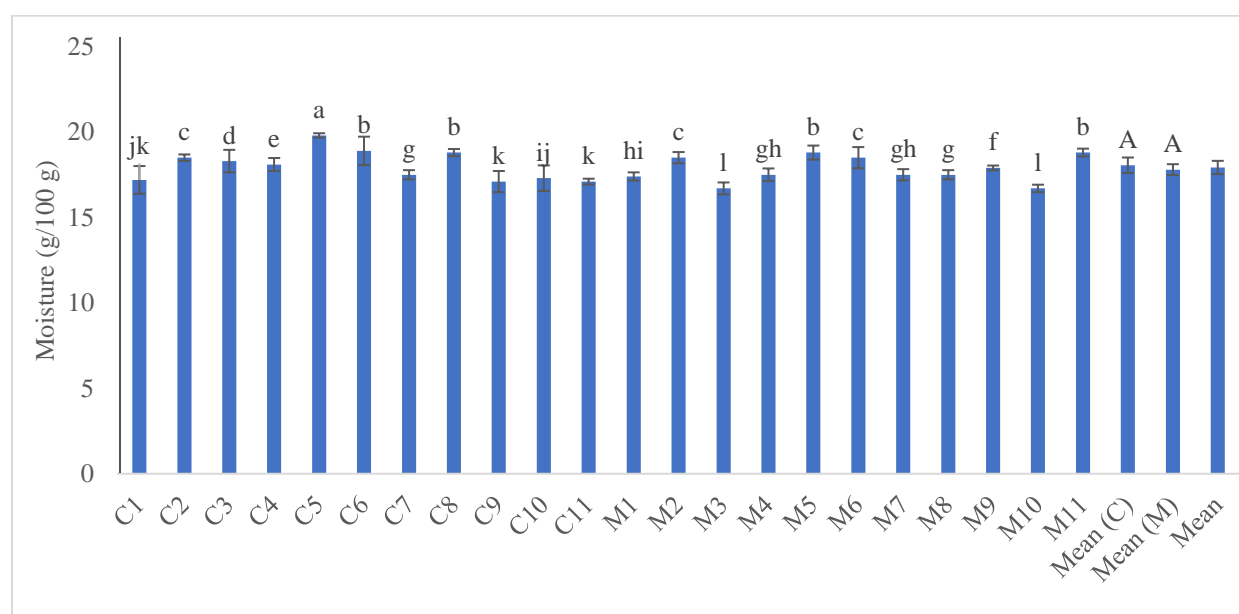


Figure 13. Moisture content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-l) above bars differ significantly (LSD test, $p < 0.05$).

According Sanz et al., (1995), honeys with moisture content less than 17.1% avoid fermentation, however, the stability of honeys with moisture content ranged between 17.1 and 20% is conditioned by a low level of yeasts and microbial content. In this study, only four samples had

moisture content of 17.1% or less, viz, C9 (17.1%), C11 (17.1%), M3 (16.7%) and M10 (16.7). On the other hand, Subramanian *et al.*, (2007) stated that the moisture content should be less than 17% for honeys to prevent fermentation. Thereby, only M3 and M10 had moisture content below 17%. The crystallization increases with the decrease of water content (Subramanian *et al.*, 2007), which may also explain that M10 was crystallized.

2.1.2. pH

Honey has an acidic nature with a pH level ranging between 3.20 and 4.50 (Da Silva *et al.*, 2016). The texture, the stability, and the shelf life of honey are affected by the pH level, and low pH usually prevents the development of microorganisms (Kumar *et al.*, 2018). As can be seen from Table 1, the pH values ranged from 3.64 to 4.59 (Figure 3) and it was significantly different

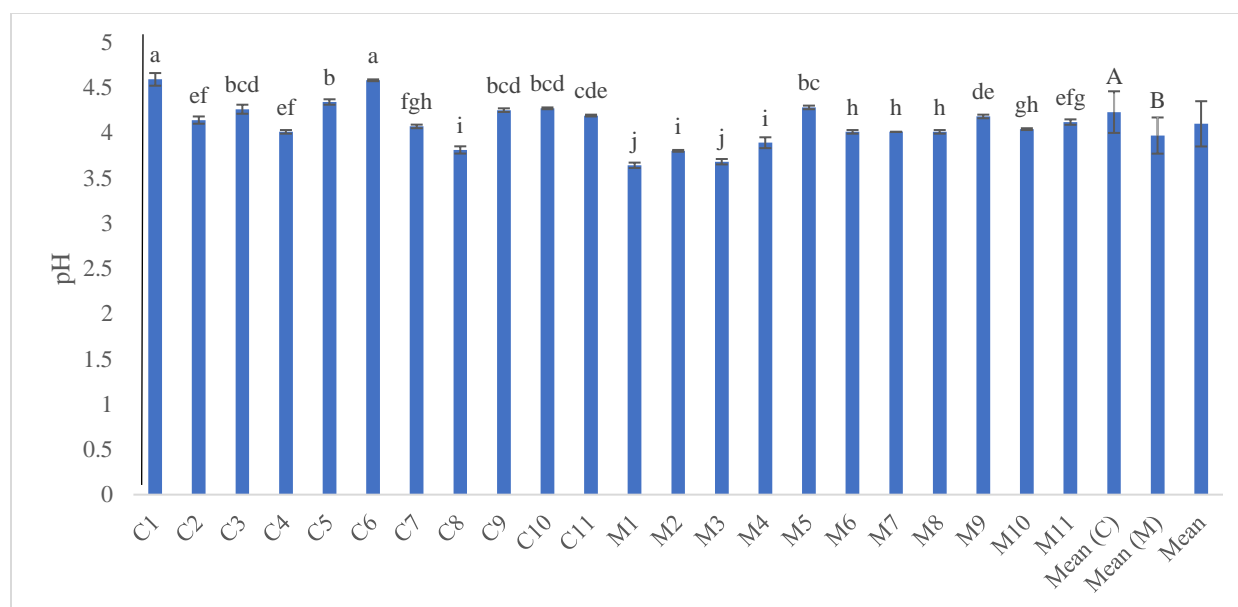


Figure 14. pH of of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (*t*-test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-j) above bars differ significantly (LSD test, $p < 0.05$).

among the samples ($p < 0.05$). The samples C1 and C6 had the highest pH values with 4.59 and 4.58, respectively, while M1 and M3 had the lowest pH values with 3.64 and 3.68, respectively. In addition, C4, M6, M7, and M8 had the same pH value with 4.01. The pH was significantly higher in coastal honeys than mountain ones with mean values of 4.22 against 3.96 ($p < 0.05$). In

addition, pH presented a significant negative correlation with the altitude ($r=-0.355$; $p<0.01$). On the other hand, Ribeiro *et al.* (2014) and Karabagias *et al.* (2014) reported different pH limits with 2.98-4.15 for Brazilian honeys and 3.40-5.31 for Greek unifloral honeys, respectively. Generally, plant source, soil, inorganic molecules, and the honey ripening process can affect the pH level of honey (Ribeiro *et al.*, 2014).

2.1.3. Free, lactonic and total acidities

In honey, organic acids represent less than 0.50% of the total composition. Nevertheless, they have a major impact on honey acidity, which influences honey flavor and boosts chemical reactions and bioactive activities (Cavia *et al.*, 2007). In addition, gluconic acid is the most important acid presented in honey, and it comes originally from the activity of glucose oxidase provided by bees through the ripening process (Karabagias *et al.*, 2014). The organic and amino acids determine the acidity of honey and they depend on the plants source (Ratiu *et al.*, 2020).

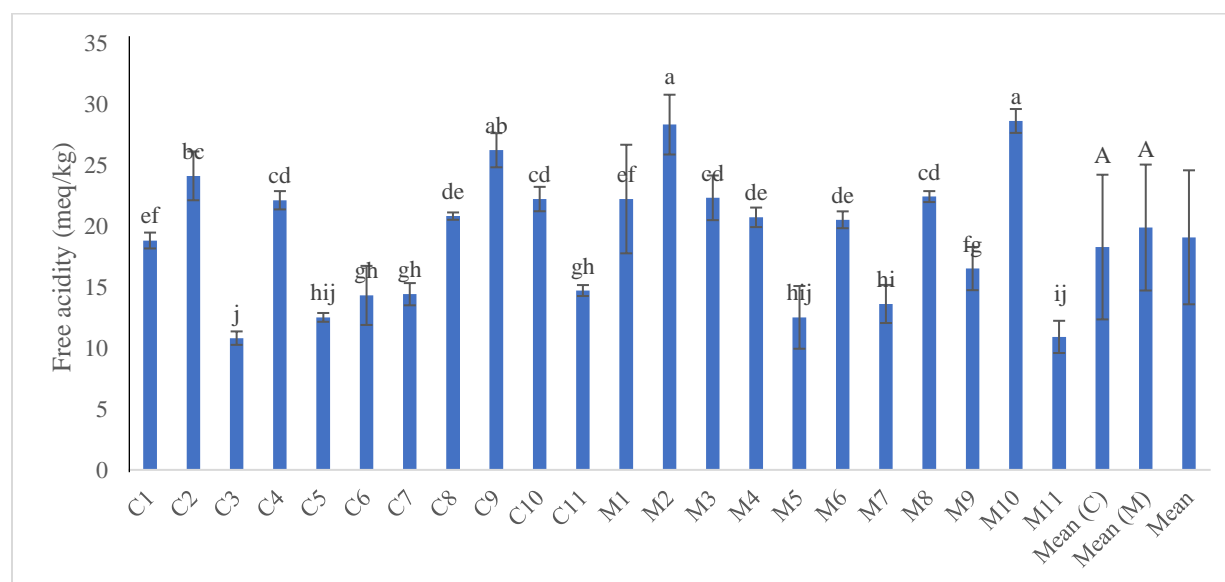


Figure 15. Free acidity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (t -test, $p<0.05$). Values C1-C11 and M1-M11 with different small letters (a-j) above bars differ significantly (LSD test, $p<0.05$).

High level of free acidity values may be a signal of the fermentation of honey by yeast. alcohol and carbon dioxide are formed during fermentation by the transformation of fructose and

glucose. In the presence of oxygen, the alcohol is converted to acetic acid and the latter increases the free acidity of the honey (Ayton et al., 2019). The free acidity ranged from 10.80 to 28.60 meq/kg (Figure 4) and differed significantly ($p < 0.05$), the samples C3 and M10 had the lowest and the highest values respectively. All samples were within the allowed limits fixed by the European Honey Commission (under 50 meq/kg) (Karabagias et al., 2014), showing the honey freshness and the absence of undesirable fermentations (Finola et al., 2007). On the other hand, Azonwade et al. (2018) reported a higher range of free acidity within 35.70 and 40.50 meq /kg for Beninese honeys, Nešović et al., (2020) found it higher in the range of 25.81 to 36.63 of honeys from Montenegro and Sajid et al., (2020) reported also higher for fresh Pakistani honeys with 33 to 46.5 meq/kg, but lower for branded Pakistani honeys with 14.16 to 16.33 meq/kg. The equilibrium between organic acids and their corresponding lactones and other mineral ions (e.g. phosphate) can be the main factor describing the level of free acidity (Finola et al., 2007).

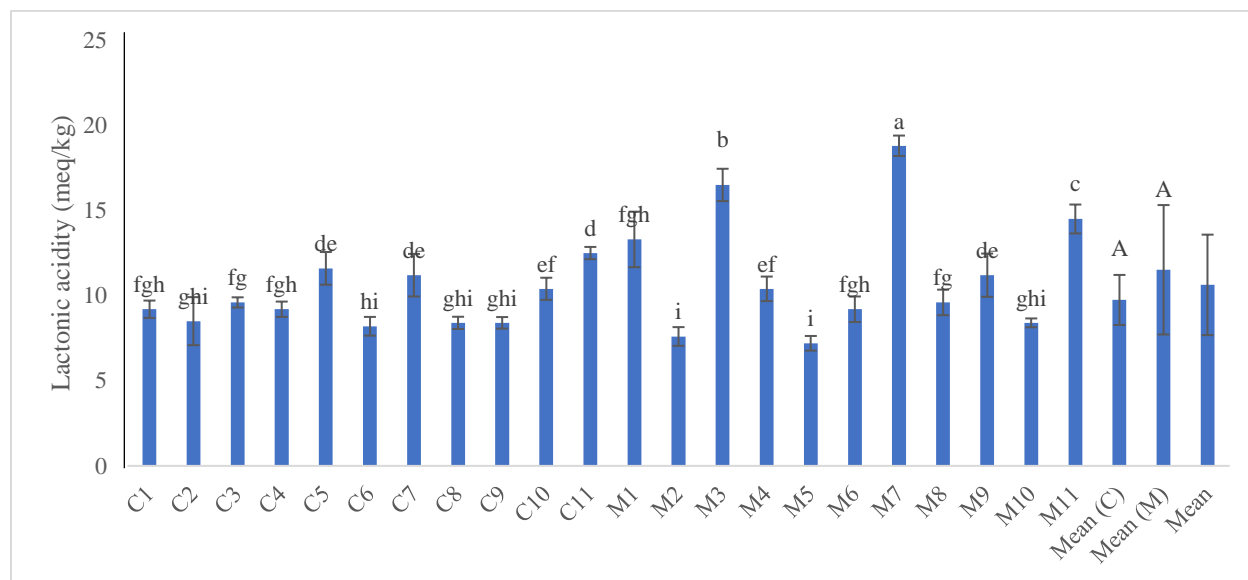


Figure 16. Lactonic acidity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-i) above bars differ significantly (LSD test, $p < 0.05$).

The hydrolysis of lactones participates in the increase of free acidity in honey, which gives interest to measure the level of lactones in honey (Bouhlali et al., 2019). In this study, the lactonic acidity ranged from 7.20 to 18.80 meq/kg, M2 had the lowest value with 8.5 meq/kg and M7 had

the highest value with 18.8 meq/kg (Figure 5) and differed significantly ($p<0.05$). Fröschle *et al.* (2018) reported different lactonic acidity range (14.5 ± 8.2 meq/kg) of *Jatropha* honey and Ahmed *et al.*, 2016 reported it lower range between 3.4 and 12.1 meq/kg of national and international honeys in Pakistan.

Finally, the total acidity ranged from 19.70 to 38.80 meq /kg and differed significantly ($p<0.05$), the samples M5 and M3 had the lowest and the highest values respectively (Figure 6). Total acidity reported ranged from 17.97 to 49.1 meq/kg by Zerrouk *et al.* (2011) for central region of Algeria, 11.94 to 58.03 meq/kg by Chakir *et al.* (2016) for Moroccan honeys and 18 to 145.50 meq/kg by Alqarni *et al.* (2016) for national and international Saudi honeys, which were higher than our results.

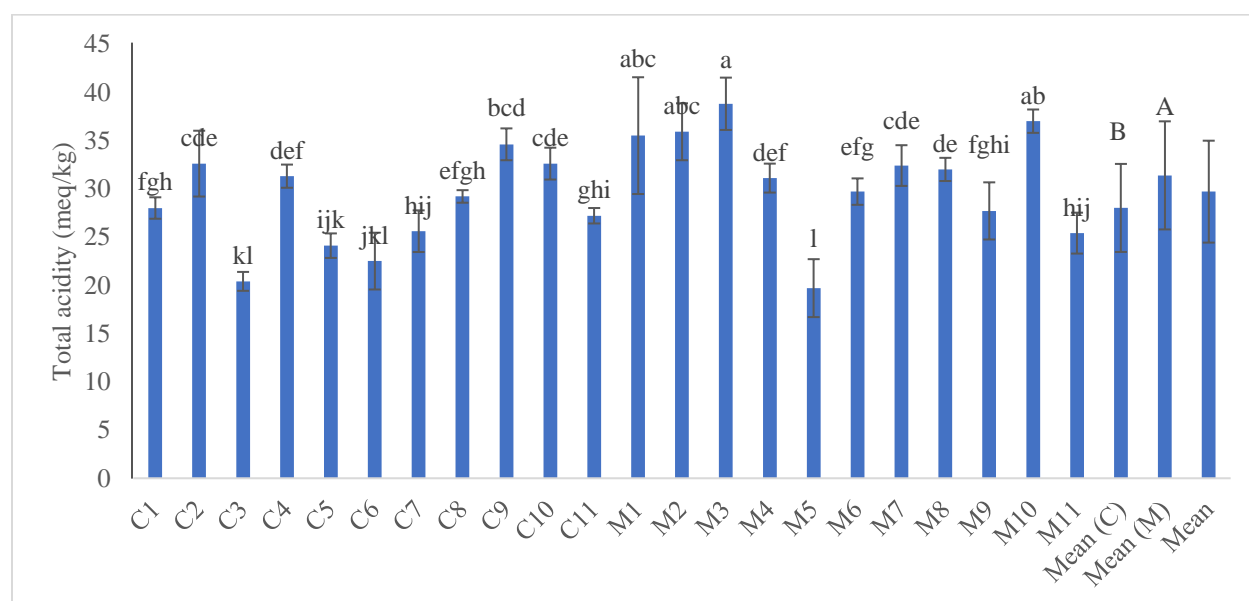


Figure 17. Total acidity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (t -test, $p<0.05$). Values C1-C11 and M1-M11 with different small letters (a-l) above bars differ significantly (LSD test, $p<0.05$).

Free acidity correlated strongly with total acidity ($r=0.856$; $p<0.01$) (Table 15), although Kumar *et al.* (2018) observed even stronger correlation between these parameters for Indian honeys ($r=0.920$). Therefore, this may indicate that the total acidity is mostly controlled by the free acidity. In addition, the total acidity and free acidity had a significant negative correlation with the pH by $r=-0.445$; $p<0.01$ and $r=-0.336$; $p<0.01$, respectively, which is well known that lower pH

causes higher acidity.

The mean value of total acidity was significantly higher in mountain honeys than coastal honeys (31.38 and 28.01 meq/kg), which can be explained by the difference of the melliferous flora between mountainous and coastal honeys. In addition, the artificial food i.e. sugar syrup may be was used more by beekeeper in mountain regions to feed honeybees, which cause the increase of acidity due to the conversion of fructose and glucose to organic acid such as acetic acid. Moreover, according to Bath and Singh (1999) the heating of honey during processing also may increase the acidity and according to Ananias et al. (2013) to explain that the high microbial content and yeasts may increase the acidity in honeys. In general, the differences of the acidity of tested honeys were due to the differences of the botanical origin, processing and the storage time and conditions.

2.1.4. Electrical conductivity (EC)

Electrical conductivity (EC) fell between 0.29 and 1.13 mS/cm, the samples M5 and M3 had the lowest and the highest values respectively (Figure 7). The results observed for the honeys differed significantly ($p<0.05$), even though, C8, M4, and M5 are not significantly different. In other studies, Can et al. (2015) and Karabagias et al. (2014) reported higher EC with 0.30 to 1.50 mS/cm for Turkish honey and 0.31 to 2.49 mS/cm for Greek unifloral honeys, respectively. On the other hand, Flores et al. (2015) found the EC of honeydew honeys was higher than 0.80 mS/cm. Indeed, mineral salt, organic acid, and protein levels are the most important factors that influence the EC of honey.

The EC negatively correlated with the altitude ($r=-0.405$; $p<0.01$) (Table 15). Likewise, EC of coastal and mountain samples were significantly different ($p<0.05$) with mean values of 0.75 mS/cm against 0.58 mS/cm. Along with the botanical origin that determines the electrical conductivity of honeys, it is possible to add three suggestions to explain these results. Firstly, electrical conductivity may be used to reveal if sugar syrup has been used to feed honeybees (Sancho et al., 2001). The adulteration of honey by artificial sugar syrups increases the electrical conductivity of honey (Salvador et al., 2019). Thus, the beekeepers in the coastal regions used sugar syrups to feed the honeybees more than in mountain regions. Secondly, low altitude regions are subjected to higher temperature coupled with the low density of vegetation compared to high altitude, may help the plants to activate their metabolisms and get more nutrients, which promote

the richness of secreted nectar. Finally, Bogdanov et al., (2007) found that most trace elements correlated significantly with electrical conductivity, these elements can be both from natural sources (soil, plants) and/or anthropogenic sources well known as potential air or soil contaminants; these results may also interpret the correlation between the EC of tested samples and the altitude, especially, that the anthropogenic action is higher in low altitude regions.

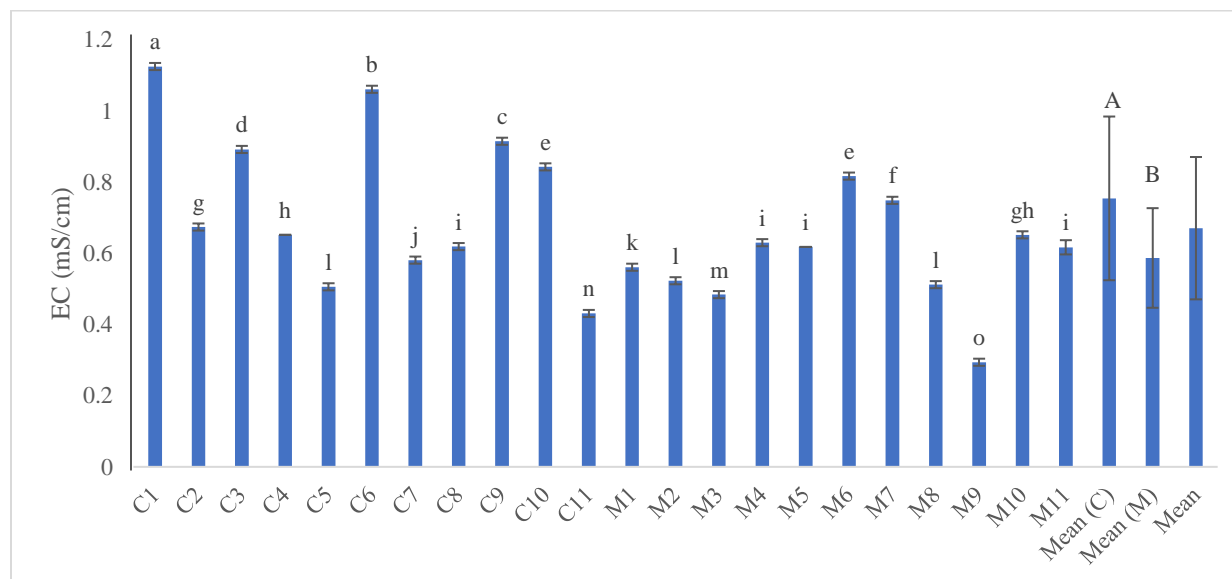


Figure 18. Electrical conductivity (EC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-o) above bars differ significantly (LSD test, $p < 0.05$).

Moreover, EC is an indicator used to distinguish floral honeys from honeydew honeys (Can et al., 2015). Generally, honeydew honeys have EC greater than 0.80 mS/cm (Bogdanov et al. 1999). Hence, tested honeys of Jijel had five coastal samples (C1 (1.13 mS/cm), C3 (0.89 mS/cm), C6 (1.06 mS/cm), C9 (0.91 mS/cm) and C10 (0.84 mS/cm)) and one mountain sample (C6 (0.82 mS/cm)) with an EC value higher than 0.80 mS/cm, indicating that these samples are more likely to be honeydew honeys or honeys of Chestnut (*Castanea*), Strawberry tree (*Arbutus unedo*), Bell Heather (*Erica*), *Eucalyptus*, Lime (*Tilia* spp), Ling Heather (*Calluna vulgaris*), Manuka or Jelly bush (*Leptospermum*) and Tea tree (*Melaleuca* spp.) or a blend of them according to the Codex Standard for Honey (2019). Therefore, *Castanea*, *Arbutus unedo*, *Erica* and *Eucalyptus* were in the list of inventoried melliferous plants of our region. According to the localizations of the tested

samples, the presence of *Eucalyptus* and *Populus nigra* (offer honeydew) may be the main factors for the high value of EC for Beni Belaid (C1) sample, *Eucalyptus*, *Quercus suber* (offers honeydew) and fruit plants that offer honeydew such as *Prunus domestica* could be the cause of the high value of EC for El Janah (C3) sample, *Eucalyptus* and fruit plants that offer honeydew could be the cause of the high value of EC for Boukartoum (C6) sample, *Quercus suber*, *Arbutus unedo*, *Erica*, *Eucalyptus* and fruit plants that offer honeydew might be the cause of the high value of EC for El Aouana (C9) and Timizer (C10) samples and *Castanea sativa*, *Arbutus unedo*, *Erica* and fruit plants that offer honeydew might be the cause of the high value of EC for Djimla (M6). The presence of contaminants in C1, C3, C6, C9, C10, could also explain the high values of EC because their proximity to the anthropogenic action.

2.1.5. Ash content

The ash content of honey has been linked with the mineral content, botanical and geographical origins (Ribeiro et al. 2014; Yücel and Sultanog 2013). Figure 8 shows that ash content of honeys from Jijel region of Algeria ranged from 0.13 to 0.83 g/100 g (the samples C5 and C6 presented the lowest and the highest values, respectively). Ouchemoukh et al. (2007) found lower ash content for Algerian honey of a different region (0.06 to 0.54 g/100 g). Da Silva et al. (2016) stated that the ash content in honey ranges from 0.02 to 1.03 g/100 g. The mean value of ash content was 0.46%, which is less than 0.50% reported by Alqarni et al. (2014) of local and imported honeys in Saudi Arabia and higher than 0.28% reported by Kahraman et al. (2010) of honeys from different regions of Turkey. The botanical origin differences could explain these results because it is the major factor that determine the ash content in honey. The ash indicates the inorganic components and it may be used to distinguish the floral origin of honey, which is (≤ 0.60 g/100 g) for blossom honeys and (≤ 1.20 g/100 g) for honeydew honeys (Kumar et al., 2018). Therefore, the five samples with ash content above 0.60 g/100g (C1 (0.81g/100 g), C3 (0.66 g/100 g), C6 (0.83g/100 g), C9 (0.62g/100 g), M6 (0.62 g/100 g)) can be determined as honeydew honeys. The ash content of C10 was 0.25 g/100 g, which means that it is mostly not a honeydew honey and the adulteration may explain its high value of the EC.

Statistically, the results showed significant differences in ash content ($p < 0.05$) and coastal honeys presented higher ash content than mountain honeys. In addition, the negative correlation ($r = -0.360$; $p < 0.01$) (Table 15) between ash content and altitude was noted. These results could be

explained by the presence of honeydew honeys in coastal regions which increase the mean value of the ash content of coastal samples compared to mountain ones. Karabagias et al., (2014) reported that the ash content can indicate the environmental pollution, this statement can explain the negative correlation of ash content with the altitude due to the high anthropogenic action in low altitude region compared to high altitude regions. Moreover, the decrease of temperature with the altitude may also interpret these results, due to the ability of temperature to urge the metabolism of plants, which increase the richness of nectar with different minerals. Finally, according to Karabagias et al., (2014) the adulteration also may increase the value of ash content.

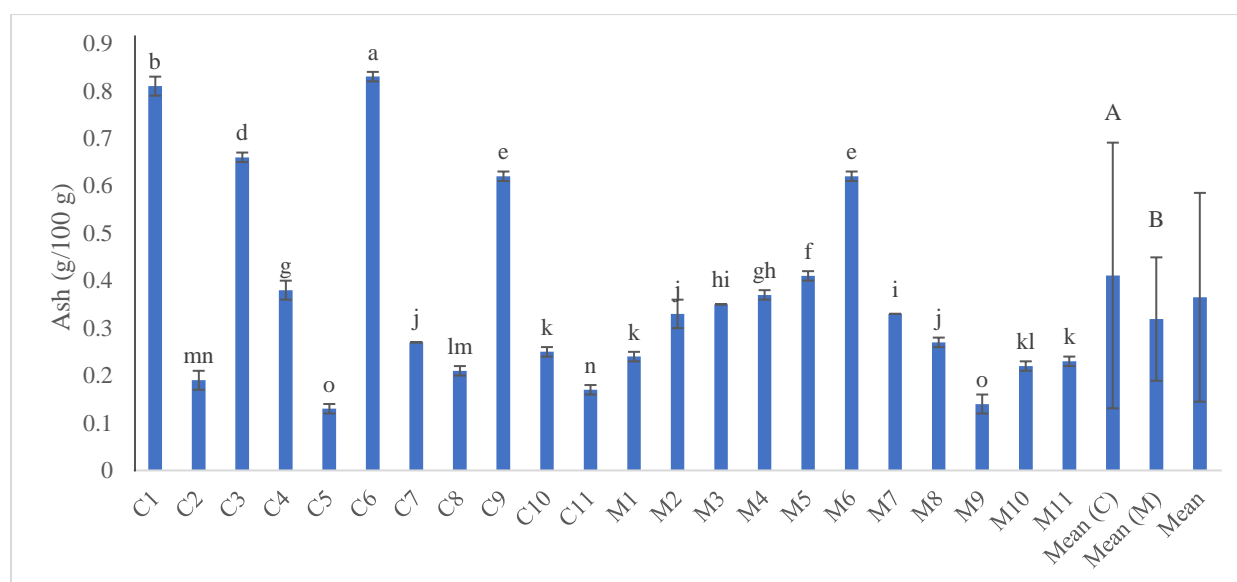


Figure 19. Ash content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (*t*-test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-o) above bars differ significantly (LSD test, $p < 0.05$).

Knowing that the electrical conductivity of honeys is governed by the ash content (Imtara et al., 2018), Da Silva et al. (2016) reported that according to Codex Alimentarius standards, the estimation of EC can replace the measurement of the ash content. In this study, the correlation coefficient between electrical conductivity and ash content was 0.885 ($p < 0.01$). Likewise, Ouchemoukh et al. (2007) and Saxena et al. (2010) obtained higher correlation for some Algerian and Indian honeys (0.92 and 0.98), respectively. Moreover, Sancho (b) et al. (1991) found the following relation: total ash (%) = 0.083 electrical conductivity - 0.092. The measurement of

electrical conductivity indicates indirectly the ash content of honey, however, high electric conductivity values do not necessarily correspond to higher amounts of ash in the honey (Baloš et al., 2018).

The quantity of ions in honey depends on the mineral content and determines the pH and electrical conductivity (Vanhanen et al., 2011). This explains the high correlation coefficient found in this study between the ash content and electrical conductivity with the pH were $r=0.524$ and $r=0.565$; $p<0.01$, respectively. On the other hand, Vanhanen et al. (2011) and Meda et al., (2005) reported higher correlation between the ash content and the pH with 0.776 and 0.77, respectively.

2.1.6. Hydroxymethylfurfural content

Hydroxymethylfurfural (HMF) is a furanic compound indicating honey freshness (Manzanares et al. 2011). It is made from dehydration of sugars in acidic conditions (caramelization) throughout heat treatment of food as an intermediate in the Maillard reaction (Pasias *et al.*, 2017). In addition, the sugar content nature, organic acids, pH, water content, and plant source affect the HMF content (Da Silva *et al.*, 2016). The HMF content increases with the storage and heat time (Fallico et al., 2004). In this study, HMF content of all honeys was under the maximum limits (40 mg/kg) approved by the Codex Standard for Honey (2019), and it ranged from 2.36 to 10.80 mg/kg (Figure 9), indicating the freshness of Jijelian honeys. The samples M7 and C2 had the lowest and the highest values, respectively. The environmental temperature, sugar type, pH, and the concentration of divalent cations in the medium control the speed of HMF formation in foods (Gregorc et al., 2020).

The statistical analysis shows that the honeys differed significantly in terms of HMF content ($p<0.05$) and the HMF content was significantly higher in coast samples than mountain samples with the mean values 7.26 mg/kg and 4.44 mg/kg, respectively. In addition, HMF presented a highly significant negative correlation with the altitude ($r=-0.510$; $p<0.01$). It could be explained by the temperature decreasing while the altitude is increasing, and more temperature produces more HMF in honey. In addition to this, generally, honey in low altitude locations is harvested earlier than in locations with a high altitude. Thereby, honey of high altitude more likely to be fresher.

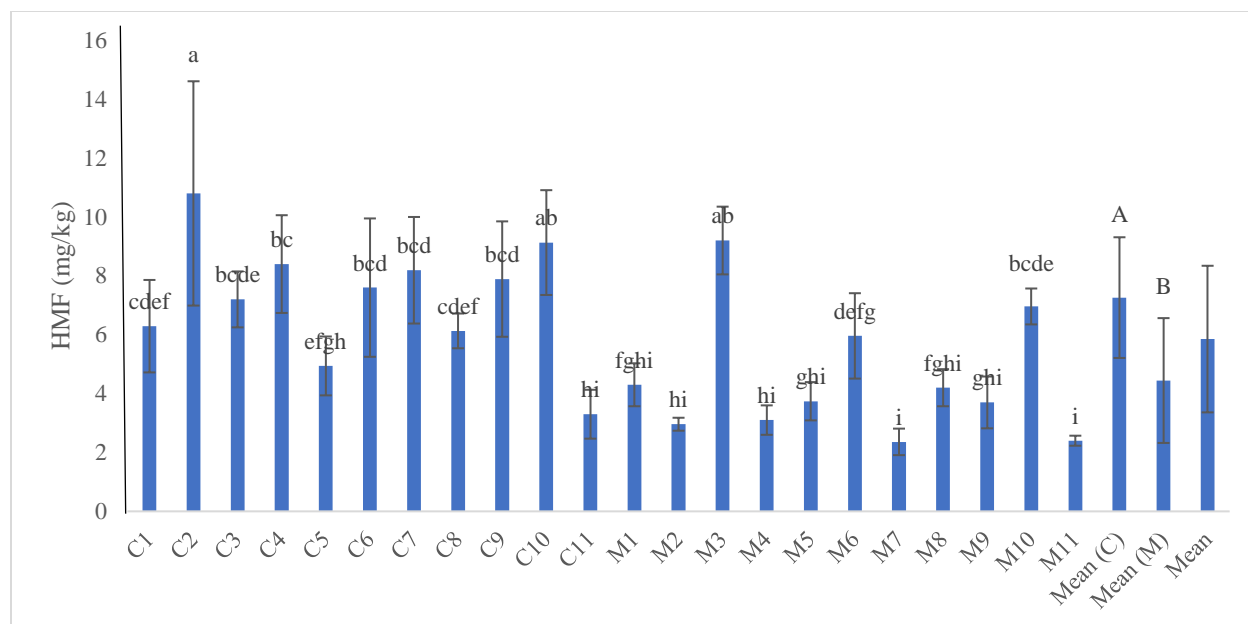


Figure 20. Hydroxymethylfurfural (HMF) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-j) above bars differ significantly (LSD test, $p < 0.05$).

HMF presented a significant correlation with the EC ($r = -0.366$; $p < 0.01$) and ash content ($r = -0.301$; $p < 0.05$). Khalil et al. (2010) reported higher correlation between HMF and free and total acidity with $r^2 = 0.786$ and 0.763 , respectively. HMF also presented a significant correlation with the free acidity ($r = -0.341$; $p < 0.01$) and total acidity ($r = -0.324$; $p < 0.01$). Belay et al. (2013) reported correlations of $r = 0.318$ between EC and HMF. HMF formation in honey is affected by the chemical properties such as pH, free acid content, total acidity, lactone content and mineral content and latter ones are strongly dependent on the botanical origin (Shapla et al., 2018). This may explain the correlation of HMF with free and total acidity, EC and Ash. The increase of these parameters is a sign of honey adulteration and the formation of HMF is activated in the acidic conditions.

2.2. Protein content

Protein represents between 0.20 and 1.60 g/100 g of honey produced by *Apis mellifera*. Both animal and vegetal sources contribute to the presence of proteins and amino acids in honey (Da Silva et al., 2016). The protein content of honeys from Jijel region of Algeria is presented in Figure 10. It ranged from 35 to 900 mg/100g and differed significantly ($p < 0.05$) (the samples M1

had the lowest value and C2 had the highest value), which was similar to the results obtained by Ouchemoukh *et al.* (2007) for Algerian honey of a different region. On the other hand, Azeredo *et al.* (2003) and Saxena *et al.* (2010) reported lower protein content for some Brazilian and Indian honeys, respectively. Abdulkhaliq and Swaileh (2017) also reported lower protein content in the range of 0.20 and 0.49% of multi-floral honey from the West Bank, Palestine. The amount of protein in honey is majorly dependent on the botanical source (Kivrak, 2017), which explains the significant differences of the protein content among the tested samples. Protein content did not present a significant correlation with the altitude.

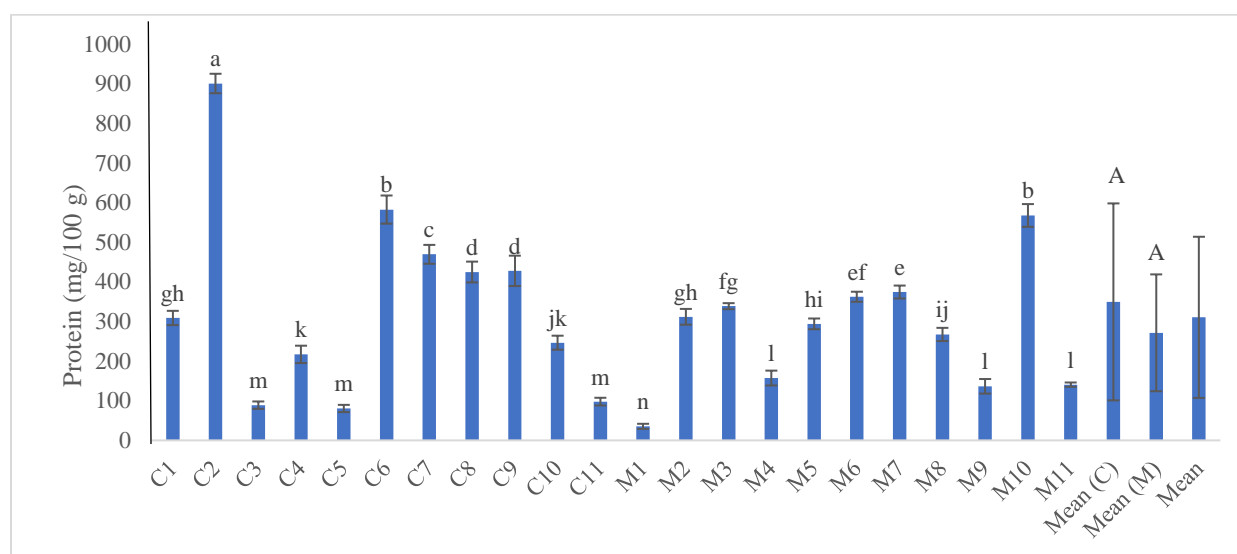


Figure 21. Protein content of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (*t*-test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-n) above bars differ significantly (LSD test, $p < 0.05$).

A high correlation coefficient of $r = 0.544$; $p < 0.01$ found between the protein content and HMF. This high correlation may be due to the formation of HMF during Maillard's reaction that happens between sugars and proteins, which means that more protein content may help to form more HMF. In addition, the protein content correlated significantly with free acidity, total acidity and EC by $r = 0.415$; $p < 0.01$, $r = 0.314$; $p < 0.05$ and $r = 0.306$; $p < 0.05$, respectively. These results are in accordance with the literature that includes the protein content in the factors that affect the electrical conductivity. On the other hand, the proteins in the tested honeys could be in an acidic nature, which could explain the correlation of the protein content with the acidity.

2.3. Color analysis

Honey color is a strong indicator of pigments' existence, like carotenoids and flavonoids, which provide a good antioxidant activity (Kek et al., 2014). Color and flavor of honey are attached to each other; while light colored honeys are mild, the darker ones present a strong flavor (Belay et al., 2015). Dark colored honeys usually have more phenolic acid derivatives but fewer flavonoids than the light colored ones (Siddiqui et al. 2016). In addition, light-colored honeys usually have low ash content, while dark-colored honeys generally have higher ash content (Alvarez-suarez et al. 2010b). The color of Jijelian honeys ranged from extra white to dark amber (Table 14). The color is arranged as follows: dark amber color dominate by ten samples (45.45%), amber by six samples (27.27%), light amber by four samples (18.18%), white color one sample and extra white one sample (4.54%). The samples C6 had the highest Pfund scale (mm) value (216) and M3 had the lowest one (16). On the other hand, Bueno-Costa et al. (2016) reported light amber (41.70%), amber (25%), and amber (33.30%) for Brazilian honeys, while Finola et al. ((2007) found white (27%), extra white (30%), white (27%), extra light amber (13%), and amber (3%) for Argentinian honeys. The differences of color among the tested samples may be due to the differences in botanical origins, which is the major factor depicts the honey color.

The color of honey is very important for commercialization, which attract the consumers and set their preferences (Da Silva et al., 2016). Generally, darker honeys tend to have more ash, nutrients, and antioxidants according to their higher correlation with bioactive compounds and different bioactive activities compared to light colored honeys. In addition, 63.63% of coastal honeys were dark amber, while only 27.27% of mountain honeys were in dark amber color. Honey color negatively correlated with the altitude ($r=-0.268$; $p<0.05$) (Table 15). These could be because the coastal honeys had more elements and nutrients than the mountainous honeys. In addition, pollution could play a role in this due to the higher anthropogenic action in coastal regions.

Color significantly correlated with the pH, ash content, EC and protein with $r=0.552$; $p<0.01$, $r=0.501$; $p<0.01$, $r=0.437$; $p<0.01$ and $r=0.323$; $p<0.01$, respectively. The correlation between the color and the ash and EC have been widely reported by many previous authors (Di Rosa et al., 2019). Escuredo et al. (2019) reported a correlation of 0.334 between color and pH. Battershall (2019) stated that the pH of honey ash is alkaline. Potassium is alkaline and it is the major mineral in honey, so more potassium leads to higher pH. Thereby, the color, ash, EC and

pH affect each other. On the other hand, it is well known that dark honeys have more nutrients such as protein and mineral compared to light honeys. The protein content affects the color of honey (Sajwani et al., 2007).

Table 14. Color analysis of tested honeys.

Sample	Color analysis (635) mm	
C1	101±10 ^g	amber
C2	122±9 ^{ef}	dark amber
C3	141±12 ^{cd}	dark amber
C4	79±9 ^{hij}	light amber
C5	52±4 ^k	light amber
C6	216±15 ^a	dark amber
C7	149±18 ^{cd}	dark amber
C8	96±8 ^{gh}	amber
C9	179±26 ^b	dark amber
C10	190±14 ^b	dark amber
C11	139±11 ^{de}	dark amber
M1	23±6 ^l	white
M2	137±11 ^{de}	dark amber
M3	16±3 ^l	extra white
M4	91±8 ^{ghi}	amber
M5	158±20 ^c	dark amber
M6	90±2 ^{ghi}	amber
M7	63±6 ^{jk}	light amber
M8	148±6 ^{cd}	dark amber
M9	76±8 ^{ij}	light amber
M10	105±7 ^{fg}	amber
M11	108±8 ^{fg}	amber
Mean (C)	134±50 ^A	Dark amber
Mean (M)	92±46 ^B	Amber
Mean	112.68	Amber

Values are presented as mean ± standard deviation (n=3). Values C1-C11 and M1-M11 with lowercase superscript differ significantly (LSD test, $p<0.05$). Means for coastal and mountain honeys (C and M, respectively) marked with different capital letters in superscript are significantly different (t -test, $p<0.05$). Mean: Total mean.

2.4. Total phenolic content

Phenolics are natural compounds known by their high importance in scientific and therapeutic research (Alvarez-Suarez *et al.*, 2010b). Their level in honey participate in profiling the antioxidant power and some sensory properties (e.g. color) (Tahir *et al.*, 2017).As can be seen

in Figure 11, total phenolic content (TPC) of Jijelian honeys was obtained in the range of 48.19 ± 6.25 to 147.50 ± 17.85 mg GAE/100 g (the samples C5 and M8 had the lowest and the highest values respectively) and it differed significantly ($p < 0.05$) among the samples. Bueno-Costa *et al.* (2016) found lower values (61.16-111.37 mg GAE/100 g) for Brazilian honeys, while Flores *et al.* (2015) reported higher values (79.50-187 mg GAE/100 g) for Spanish honeydew honeys. Floral and geo-graphical origins are the major factors determine the level of phenolics content of honey (Čanadanović-Bruneta *et al.* 2014).

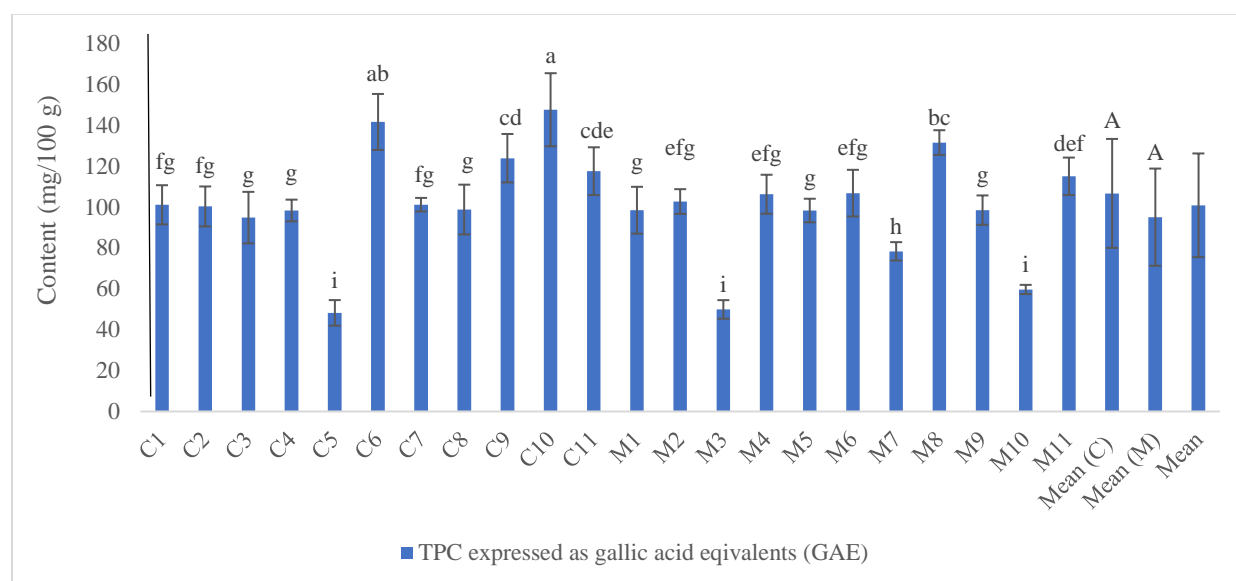


Figure 22. Total phenolic content (TPC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-i) above bars differ significantly (LSD test, $p < 0.05$).

TPC did not present a significant correlation with the altitude and there was no significant difference between coastal and mountain honeys. In honey, the level of phenolic content is based on food source, geo-graphical origin, processing, handling, and storage (Flores *et al.*, 2015). From these results, we can deduce that there is no geographical pattern affects the amount of phenols present in honey. Whereas, TPC had a high significant correlation with color with $r = 0.738$; $p < 0.01$. Al-Farsi *et al.* (2018) declared that polyphenol has been widely mentioned and known to have a high correlation with honey color and reported a higher correlation between color and phenolics (0.974) of Omani honeys. Likewise, Pontis *et al.* (2014) reported higher correlation between

phenolics and color i.e. 0.967 of Brazilian honeys. In addition, it is well known that the phenols get a brownish color if they were subjected to oxidation, so more oxidized phenols make honey darker. Moniruzzaman et al. (2014) suggested that the phenolic compounds are the provider for color pigments of honey. On the other hand, TPC had a high significant correlation with ash, EC and pH with $r=0.455$; $p<0.01$, $r=0.339$; $p<0.01$ and $r=0.314$; $p<0.05$, respectively. Phenolics can be neutral or have an acidic nature (Nakanishi et al., 2013). The correlation between phenols and pH could be due to the nature of phenolics as weak acids. Darker honeys generally have higher minerals and phenolics, which may explain the correlations between phenolics and ash and EC. In addition, it was documented in many previous studies that honeydew honey, which generally have more mineral contents, have higher phenolic content than blossom honeys.

2.5. Total flavonoid content

Flavonoids have a substantial contribution to the antioxidant properties of honey, and they are described as the most important functional compounds of honey (Da Silva *et al.*, 2016). The total flavonoid content (TFC) of honeys from Jijel varied between 5.54 ± 0.33 and 46.88 ± 1.67 mg QE/100 g (the samples M3 and M8 had the lowest and the highest values, respectively) (Figure 12), and it differed significantly ($p<0.05$). Chaikham *et al.* (2016) obtained higher TFC values ranging between 31.52 and 60.73 mg QE/100 g for Thai monofloral honeys, whereas Tenore *et al.* (2012) obtained lower values ranging between 6.85 and 23.17 mg QE/100 g for Italian monofloral honeys. On the other hand, Flores *et al.* (2015) reported lower values in the range of 6.60 and 13.10 mg QE/100 g for Spanish honeydew honeys. Several researchers have already reported that the floral source affects the flavonoid content of honey, and the environmental and climatic conditions depict the nectar composition of melliferous flora (De Sousa *et al.*, 2016). Flores et al. (2015) stated that honey is considered a natural flavonoid food source.

Furthermore, TFC did not present a significant correlation with the altitude and there was no significant difference between coastal and mountain honeys. These results might indicate the absence of any geographical pattern that may affect the amount of flavonoids present in honey. On the other hand, TPC had a high significant correlation with TPC and color with $r=0.802$; $p<0.01$ and $r=0.770$; $p<0.01$, respectively. Therefore, we suggest that the flavonoids were the main phenolics responsible of conferring the color pigments to honey. Sant'Ana et al. (2014) reported lower correlation between TFC and color and TPC of Brazilian honeys with 0.6 and 0.5,

respectively. On the other hand, Khalil et al. (2012) reported higher correlation between TFC and color and TPC for other Algerian honeys with 0.968 and 0.776, respectively. Likewise, Moniruzzaman et al. (2014) found higher correlation between TFC and color i.e. 0.926. Many other authors have reported the strong correlation between TFC, TPC and color. Additionally, TFC had a significant correlation with ash, EC and pH by $r=0.391$; $p<0.01$, $r=0.335$; $p<0.01$ and $r=0.371$; $p<0.01$, respectively. These results can be explained the same way as for TPC.

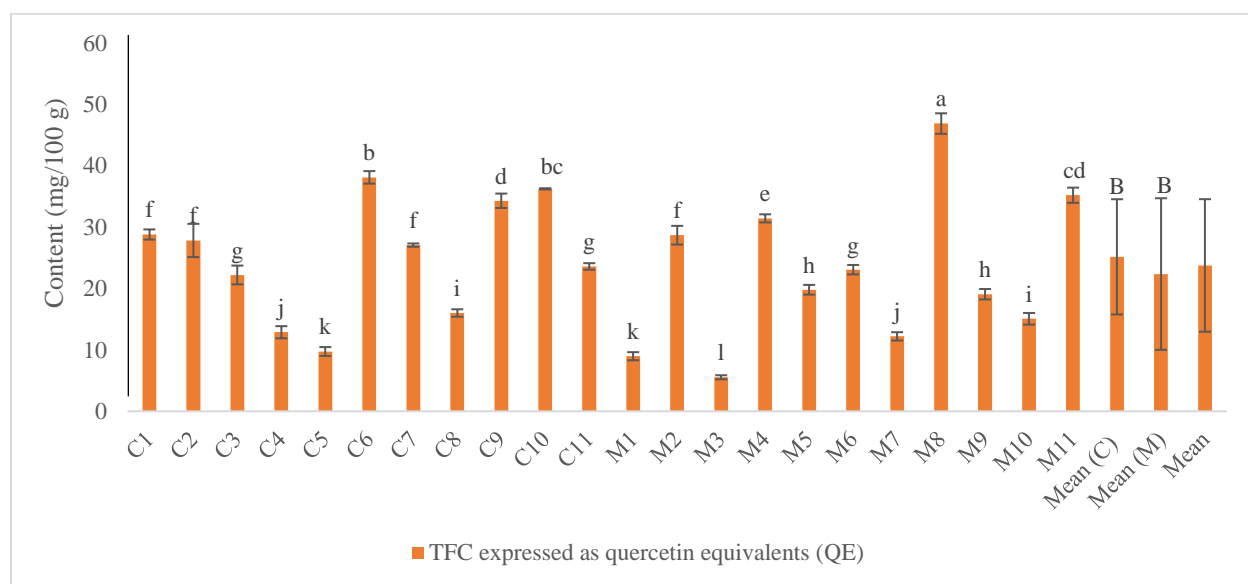


Figure 23. Total flavonoid content (TFC) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (t -test, $p<0.05$). Values C1-C11 and M1-M11 with different small letters (a-l) above bars differ significantly (LSD test, $p<0.05$).

2.6. DPPH radical scavenging activity

Recently, honeybees and honey products have been utilized as natural antioxidant sources. In addition, to assess the bioactive features of honey, the antioxidant activity is considered among the most valuable methods (Tahir et al., 2017), which is largely evaluated as DPPH radicals scavenging activity (Liu et al., 2013). The principal antioxidants in honey are flavonoids and simple phenolic derivatives (Al-Farsi et al., 2018). The Figure 13 exhibits the DPPH radical scavenging activity (RSA) expressed as IC_{50} of tested honeys, which differed significantly ($p<0.05$) between 4.2 ± 0.33 and 17.92 ± 0.87 mg/mL (the samples C10 and C5 had the lowest and the highest values respectively). Lower IC_{50} means better radical scavenging activity. These results

could be due to differences in botanical origins of samples. Escuredo et al. (2013) and Kishore et al. (2011) reported similar values ranged between 8.60-17.80 mg/mL and 5.8-10.86 mg/ml for Spanish and Malaysian honeys, respectively. However, Meda et al. (2005) and Beretta et al. (2005) reported higher RSA with IC₅₀ in ranges 1.63 to 29.13 mg/mL for Burkinabe honeys and 1.63-45.45 mg/mL for Italian honeys, respectively.

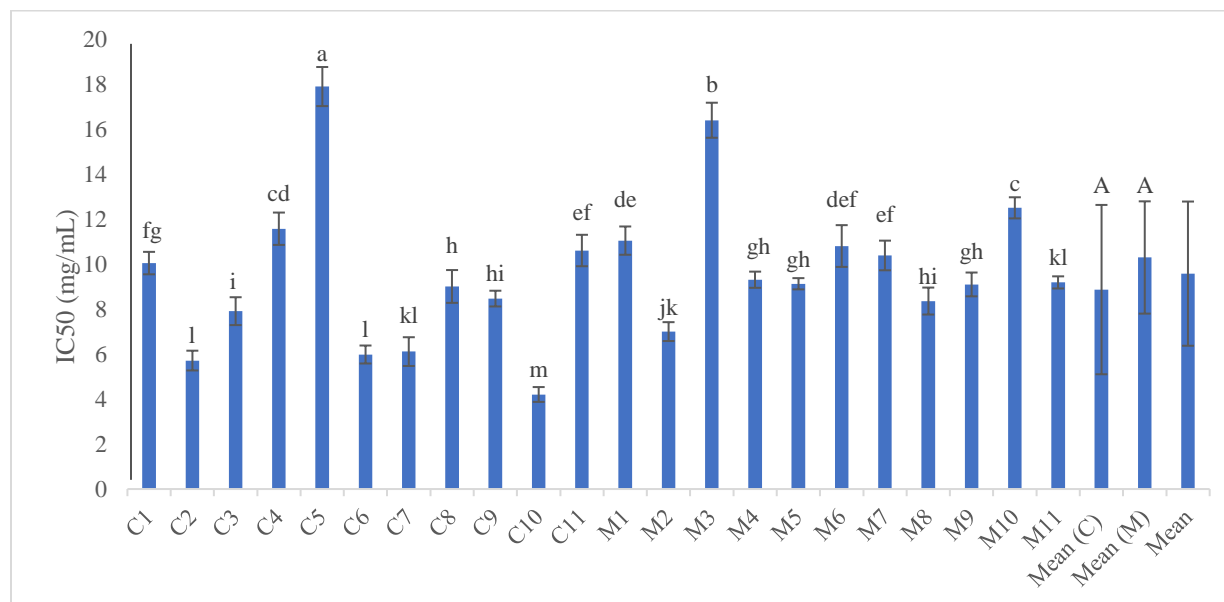


Figure 24. DPPH radicals scavenging activity of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Capital letter (A) above these bars indicate no statistically significant differences between the two groups (*t*-test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-l) above bars differ significantly (LSD test, $p < 0.05$).

RSA did not present a significant correlation with the altitude and there was no significant difference between coastal and mountain honeys. These results might indicate the absence of any geographical pattern that may affect the RSA of tested samples. Whereas, RSA had a high significant negative correlation with TPC, TFC and color by $r = -0.725$; $p < 0.01$, $r = -0.714$; $p < 0.01$ and $r = -0.691$; $p < 0.01$, respectively. These results are in accordance with what many previous authors have reported and they considered the phenolics as the most important compounds that determine the antioxidant properties of honey. Thereby, Al et al., (2009) reported a higher correlation between RSA and TPC and TFC of different floral origin honeys from Romania with $r^2 = 0.94$ and $r^2 = 0.83$, respectively. Rababah et al., (2014) also reported higher negative correlation between RSA and flavonoids with $R = -0.868$ of honeys from different Mediterranean floral sources

in Jordan. The high correlation between the antioxidant properties and phenolics in honey have been revealed by other authors (Bridi et al., 2017). Moreover, RSA had a significant negative correlation with ash, protein and EC by $r=-0.315$; $p<0.01$, $r=0.286$; $p<0.05$ and $r=-0.297$; $p<0.05$, respectively. Khalil et al. (2012) found higher correlation between RSA and proteins with 0.94 for other Algerian honeys. Phenolic acids and flavonoids, certain enzymes (glucose oxidase and catalase), ascorbic acid, proteins, and carotenoids in honey have been demonstrated to have antioxidant activity (Lokossou et al., 2017).

2.7. Reducing power (RP)

The reducing power is widely known as a strong criterion of antioxidant capacity (Küçük et al., 2007). The absorbance values of RP assay differed significantly ($p < 0.05$) between 0.11 and 0.47, while the samples M3 and C9 had the lowest and the highest values, respectively (Figure 14). Küçük et al. (2007) reported that it varied from 0.11 to 0.78 for three concentrations (1, 5 and 10%) of Turkish honeys, while Saxena et al. (2010) reported that it ranged between 0.38 and 0.59 for 10% (v/v) of Indian honeys. The reducing power may differ due to the presence of different types of phenolic compounds, non-phenolic compounds (vitamins and amino acids) and other molecules such as enzymes (glucose oxidase and catalase) (Mouhoubi-Tafnine et al., 2016).

Moreover, coastal honeys presented better reducing power than mountain honeys (0.35 against 0.29) and negatively correlated with the altitude ($r=-0.265$; $p<0.05$) (Table 15). These results could be explained by the higher mineral content of coastal honeys compared to the mountain honeys, especially, that RP had also a significant correlation with the ash content ($r=0.360$; $p<0.01$) and EC ($r=0.323$; $p<0.01$). In addition, RP had a high significant correlation with TPC, RSA ($1/IC_{50}$), TFC and color by $r=0.665$; $p<0.01$, $r=0.663$; $p<0.01$, $r=0.615$; $p<0.01$ and $r=0.609$; $p<0.01$, respectively. These results may indicate that the phenolics are the major factor responsible for the reducing power of honey. The correlation between RP and phenolics were already demonstrated by previous studies. Moniruzzaman et al. (2013) found strong correlation between FRAP and DPPH (0.850), phenolics (0.780), flavonoids (0.595) and color (0.557) of Malaysian acacia honey. Likewise, Savatović et al. (2011) reported higher correlation between the reducing power ($1/EC_{50}$) and total phenolics (0.9003) and total flavonoids (0.9984) of Serbian floral honeys. Attanzio et al. (2016) obtained also higher correlation between FRAP and phenolics (0.883), flavonoids (0.956) and DPPH (0.819) of Sicilian honeys.

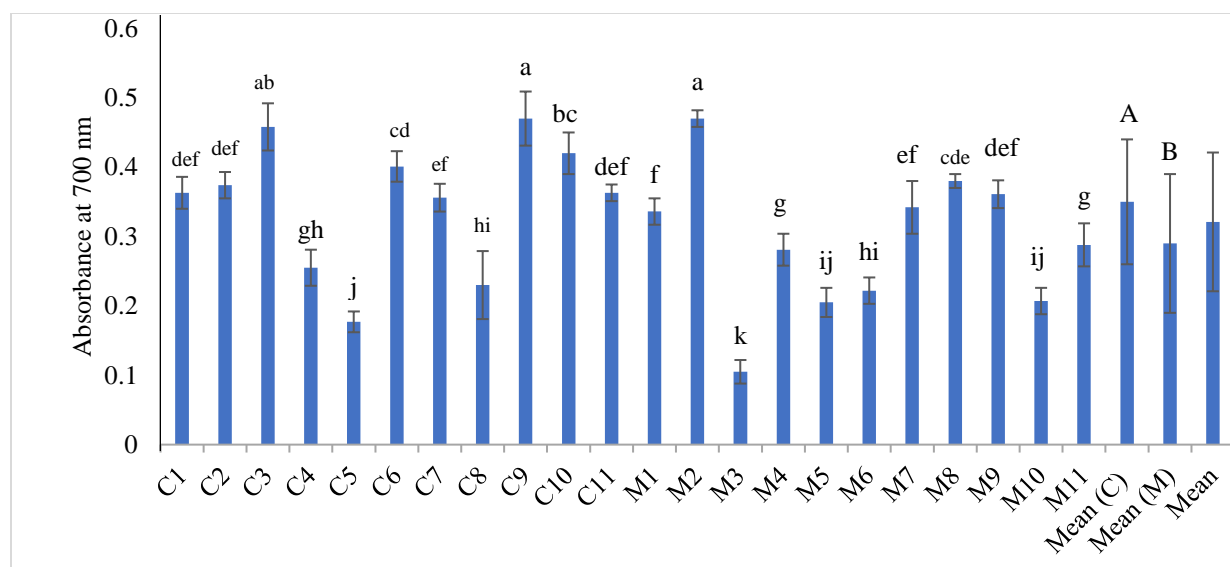


Figure 25. Reducing power (RP) of honeys from coastal (C1-C11) and mountain (M1-M11) regions of Jijel (Algeria). Bars Mean (C), Mean (M) and Mean present mean values for coastal, mountain and total honeys, respectively. Different capital letters (A-B) above these bars indicate significant differences between the two groups (t -test, $p < 0.05$). Values C1-C11 and M1-M11 with different small letters (a-k) above bars differ significantly (LSD test, $p < 0.05$).

Table 15. Pearson correlation coefficients among parameters.

	Altitude	Moisture	pH	EC	Ash	Free acidity	Lactonic acidity	Total acidity	Pfund scale	HMF	Protein	TPC	TFC	RSA IC ₅₀	RP	Methylparathion	Coumaphos	Fenitrothion
Altitude	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Moisture	-0.168	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
pH	-0.355**	0.255*	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
EC	-0.405**	0.003	0.565**	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Ash	-0.360**	-0.076	0.524**	0.885**	1	-	-	-	-	-	-	-	-	-	-	-	-	-
Free acidity	0.045	-0.391**	-0.336**	-0.011	0.005	1	-	-	-	-	-	-	-	-	-	-	-	-
Lactonic acidity	0.286*	-0.228	-0.187	-0.230	-0.239	-0.307*	1	-	-	-	-	-	-	-	-	-	-	-
Total acidity	0.202	-0.524**	-0.445**	-0.136	-0.124	0.856**	0.230	1	-	-	-	-	-	-	-	-	-	-
Pfund scale	-0.268*	0.102	0.552**	0.437**	0.501**	0.038	-0.415**	-0.187	1	-	-	-	-	-	-	-	-	-
HMF	-0.510**	-0.118	0.176	0.341**	0.324**	0.366**	-0.135	0.301*	0.234	1	-	-	-	-	-	-	-	-
Protein	-0.138	-0.012	0.108	0.306*	0.104	0.415**	-0.204	0.314*	0.323**	0.544**	1	-	-	-	-	-	-	-
TPC	-0.150	0.015	0.314*	0.339**	0.455**	0.062	-0.270*	-0.083	0.738**	0.108	0.053	1	-	-	-	-	-	-
TFC	-0.010	0.019	0.371**	0.335**	0.391**	0.104	-0.278*	-0.045	0.770**	0.024	0.185	0.802**	1	-	-	-	-	-
RSA IC ₅₀	0.093	-0.038	-0.202	-0.297*	-0.315**	0.020	0.321**	0.195	-0.691**	-0.038	-0.286*	-0.725**	-0.714**	1	-	-	-	-
RP	-0.265*	-0.099	0.294*	0.323**	0.360**	0.093	-0.203	-0.016	0.615**	0.067	0.058	0.665**	0.609**	-0.663**	1	-	-	-
Methylparathion	-0.554**	0.028	0.278*	0.408**	0.247*	0.103	-0.244*	-0.028	0.176	0.294*	0.029	-0.031	-0.166	0.101	0.037	1	-	-
Coumaphos	-0.538**	0.097	0.227	0.258*	0.147	0.155	-0.261*	0.016	-0.143	0.320**	0.041	-0.051	-0.198	0.129	-0.109	0.915**	1	-
Fenitrothion	-0.485**	0.052	0.146	0.349**	0.227	0.170	-0.285*	0.019	-0.105	0.240	0.037	0.049	-0.158	0.053	-0.071	0.891**	0.933**	1

** Correlation is significant at the 0.01 level. * Correlation is significant at the 0.05 level. EC: electrical conductivity, HMF: Hydroxymethylfurfural, TPC: Total phenolic content, TFC: Total flavonoid content, RSA: DPPH radical scavenging activity, RP: Reducing power.

Color intensity, TPC, TFC, RSA (1/IC₅₀) and RP had high correlations ranging from $r = 0.609$ to $r = 0.802$ (Table 15) (TFC and RP had the lowest correlation, while TPC and TFC had the highest correlation). Beretta *et al.* (2005) found that the correlations between color, phenol content, DPPH 1/IC₅₀ and FRAP of honey from different origin were ranging between $r = 0.884$

and $r = 0.993$. In addition, Alvarez-Suarez *et al.* (2010b) found that the correlations between color, TPC, TFC, TEAC and FRAP of monofloral Cuban honeys were ranging between $r = 0.83$ and $r = 0.97$. Furthermore, Ferreira *et al.* (2009) reported that dark honey had higher phenolics content, DPPH scavenging activity and reducing power than amber and light honeys.

2.8. Antibacterial activity

Table 16. Antibacterial activity of tested samples.

Sample	I of ESC 100% (mm)	I of ESC 50% (mm)	I of ESC 25% (mm)	I of ST 100% (mm)	I of ST 50% (mm)	I of ST 25% (mm)	I of PS 100% (mm)	I of PS 50% (mm)	I of PS 25% (mm)
C1	9± 1	9± 0.57	9± 0.57	0	0	0	0	0	0
C2	16± 0.57	0	0	8± 0.57	0	0	11± 1	0	0
C3	1± 1.730	10± 1	10± 0	0	0	0	0	0	0
C4	8± 0	8± 0.57	0	0	0	0	0	0	0
C5	10± 0.57	0	0	0	0	0	0	0	0
C6	10± 1	1± 0	9± 1	0	0	0	0	0	0
C7	0	0	0	0	0	0	0	0	0
C8	13± 1.73	0	0	10± 1	0	0	10± 0	0	0
C9	8± 1	0	0	0	0	0	0	0	0
C10	10± 0.57	8± 0.57	7± 0	0	0	0	0	0	0
C11	7± 0	8± 1	8± 0.57	0	0	0	0	0	0
M1	10± 0.57	7± 0	0	9± 1.73	0	0	0	0	0
M2	8± 0	8± 1	0	0	0	0	0	0	0
M3	10± 1	0	0	0	0	0	0	0	0
M4	9± 1	9± 1.73	8± 1	0	0	0	0	0	0
M5	9± 0.57	8± 0	9± 0	0	0	0	0	0	0
M6	10± 0	1± 0	8± 0.57	0	0	0	0	0	0
M7	0	0	0	0	0	0	0	0	0
M8	12± 1	0	0	0	0	0	0	0	0
M9	12± 1.73	0	0	8± 0	0	0	0	0	0
M10	9± 0	8± 1	8± 1	0	0	0	0	0	0
M11	11± 1.73	10± 1.73	9± 1.73	0	0	0	0	0	0

I: inhibition; ESC: *Escherichia coli*; ST: *Staphylococcus aureus*; PS: *Pseudomonas aeruginosa*

Honey is increasingly valued for its antibacterial activity especially with the rise in prevalence of antibiotic resistant bacteria. The antibacterial activity of honey is mostly depicted by the collective effect of acidity, osmolarity, hydrogen peroxide activity, and phenolic compound content (Das et al. 2015; Molan 1992). It can be due to its ability to generate hydrogen peroxide

by the bee-derived enzyme glucose oxidase (Almasaudi et al. 2017). The 100% concentration of Jijelian honeys had better antibacterial activity than 50% and 25 % concentrations for the three strains. However, 20 samples (90.9%) of Jijelian honeys had antibacterial activity against *E. coli*, 4 samples (18.18%) against *S. aureus*, and only 2 samples (9.09%) against *P. aeruginosa* (Table 4). Therefore, Jijelian honeys were more efficient against *E. coli*, than *S. aureus* and *P. aeruginosa*. Ten samples of each coast and mountain honeys presented antibacterial activity against *E. coli* while only C2, C8, M1, and M9 presented antibacterial activity against *S. aureus*. However, only two coast honeys (C2, C8) had antibacterial activity against *P. aeruginosa*. The results showed significant differences in antibacterial activity ($p < 0.05$) and only two coast samples (C2 and C8) presented antibacterial activity against the three strains of bacteria. Molan (1992) mentioned that honey have antibacterial activity against *E. coli*, *S. aureus*, and *P. aeruginosa*. Our results showed that *E. coli* was the most sensitive microorganism to Jijelian honeys and *P. aeruginosa* was the most resistant one. On the other hand, Alvarez-Suarez et al. (2010b) and Bueno-costa et al. (2016) reported that *S. aureus* was the most sensitive microorganism toward Cuban and Brazilian honeys respectively.

2.9. Pesticides

2.9.1. Pesticides analysis

Honey beekeeping on farms is the most common on the market which is the most vulnerable to pollution because of its presence near sources of insecticides, unlike those found naturally in the mountains and valleys (Zhang et al. 2018). The contaminated honey poses a potential risk to human health because of subacute and chronic toxicity of contaminations (Bogdanov, 2006). Insecticides take two main routes to reach their target. They can cross the integument of the melliferous or when they move on the residues of the product deposited on the plants (Bogdanov 2006). They are also ingested during the consumption of contaminated nectar at the bottom of the corolla (Jablonski et al. 1995). This contamination is stronger as the insecticide can have endotherapeutic properties; thus, penetrating easily into the vessels conducting sap (Colin et al. 2004). There is an insidious mode of contamination: the transport of polluted food to the nest by the foragers, which will be used to feed, either adult congeners or larvae (Fries et al. 2001). Pesticides contamination has been studied by several researchers in various honey samples collected in different regions of the world (Rissato et al. 2007; Abdallah et al. 2017; López et al.

2014). In this study, the chromatographic analyses did not show a high contamination level of pesticides, only organophosphorus (OPs) pesticides have been found exceeding the allowed limits in some samples (Table 17). The OPs pesticides are widely used in the agro-food industry for phytosanitary purposes in crops (Martinez-Toledo et al. 1992). The main OPs detected in our collected honey samples are Coumaphos, Methyl parathion and Fenitrothion. New Maximum Residue Levels (MRLs) for certain pesticides in honey, ranging from 10 to 50 ng/g, have been set since September 2008 by the European Commission (Bargańska et al. 2013). Albero et al. (2004), López et al. (2014) and Valdovinos-Flores et al. (2017) reported the presence of Coumaphos, Methyl parathion and Fenitrothion in honey.

Table 17. Summary table of the results of the analysis of the optically active organophosphorus pesticides by GC chromatography of the honey samples collected in different region of Jijel, east of Algeria.

Samples	Organophosphorus pesticides in honey ng/g		
	Methyl parathion	Coumaphos	Fenitrothion
Beni Belaid (C1)	71	55	62
Oued Zhour (C2)	27	22	8
El Janah (C3)	18	13	6
Achouat (C4)	56	72	67
El Balouta (C5)	15	20	7
Boukhartoum(C6)	3	10	12
Jijel (C7)	1	8	3
El Kennar (C8)	33	43	64
El Aouana (C9)	17	9	19
Timizer (C10)	7	15	12
Ziama Mansouriah (C11)	5	8	1
El Milia (M1)	21	09	16
Ouled Yahia (M2)	1	7	5
Ghebala (M3)	2	7	2
Bordj Thar (M4)	0	9	3
Oudjana (M5)	0	5	3
Djimla (M6)	4	4	5
Ouled Askeur (M7)	4	3	3
Taksana (M8)	2	6	1
Selma (M9)	3	8	2
Teyana (M10)	0	9	3
Erraguen (M11)	1	1	2

The mean value of the three pesticides were significantly higher in coastal samples than mountain samples ($p<0.05$). In addition, the altitude had a significant negative correlation with Methyl parathion, Coumaphos and Fenitrothion by $r=-0.554$, $r=-0.538$ and -0.485 ($p<0.05$), respectively (Table 15). These results could be due to the anthropogenic action that is higher in low altitude regions than high altitude region.

2.9.2. Organophosphorus pesticides photodegradation

The results of the retention times were shown in the chromatogram (Figure 26). The chromatogram of the standard revealed the presence of three peaks with the retention time of 21.2, 21.3 and 21.9 min attributed to Methyl parathion, Coumaphos and Fenitrothion organophosphorus pesticides, respectively. The solution containing the mixture of OPs pesticide extract was injected using, for the separation, a proprietary mobile phase HP-608 polysiloxane. Standards corresponding to the assumed analyte matrix were injected in order to compensate for the influence of the matrix on the proportion of the compounds. We had repeated injections of the standard corresponding to the matrix of our honey samples. The difference between the retention times of the analytes in the standard and the presumed OPs' peaks in the honey samples is less than 0.1 %. The chromatogram of our honey samples (Figure 26) had only three characteristic peaks because the labeled compounds had the same chromatographic behavior as the standards molecules which contains the Methyl parathion, Coumaphos and Fenitrothion with the same retention times, identical on the chromatogram. All the OPs compounds present in the honey were separated under these applied analytical conditions, which were well optimized and had clearly allowed studying their photodegradation kinetics in the present work.

The solar energy is available throughout the year with varying intensities and can constitute an alternative source that can be used especially in developing countries where the solar constant exceeds 1 kW m^{-2} (Lahmar et al. 2015). Indeed, this renewable and clean energy source is free and inexhaustible and is by far the most abundant and attractive energy on Earth. Therefore, the photocatalytic treatment of organophosphates presents itself as a technology of choice for pollution control compared to other existing conventional methods because it is a powerful, simple and economical system. These attractive characteristics have generated a great interest on the part of researchers for the understanding, optimization and industrial application of this process. Honey is considered contaminated if the pesticide residue levels exceed the MRL. Therefore, it is

necessary to establish reliable, efficient and economical methods for the elimination of pesticides in honey (Malhat et al. 2015). Numerous studies have revealed behaviors of pesticides under direct excitation (Yuan et al. 2014; Bahena et al. 2006) and made it possible to better understand these processes which participate amply to the degradation of the pesticides under this action and by time to convert them to a variety of less harmful forms.

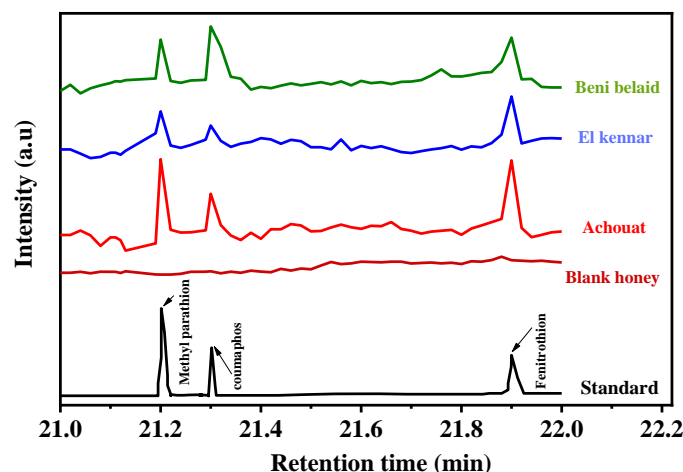


Figure 26. Preliminary chromatographic analysis (GC) of honey samples on the HP-608 polysiloxane capillary column.

The recovery efficiency of the extraction was detected before the solar light irradiation experiments. Preliminary tests using OPs' standards revealed that our honey samples have three different concentrations of the majority OPs in 3 samples (Table 17) exceeding the MRL limits that were extracted according to the method described before. Dark-controlled experiments were conducted with negligible loss of OPs (> 96% recovered) which is due to the low loss process during the experiment by hydrolysis, desorption and volatilization. On the other hand, we recovered 68% of Fenitrothion in the dark which is normal because of its volatility according to the literature, implying that the hydrolysis of OPs was not significant during the experimental period. Equation 1 was used to calculate the residual percentage (D):

$$D = \frac{(C_0 - C_t)}{C_0} \times 100 \quad (1)$$

where C_0 and C_t are the concentrations of OPs before and after photodegradation, respectively.

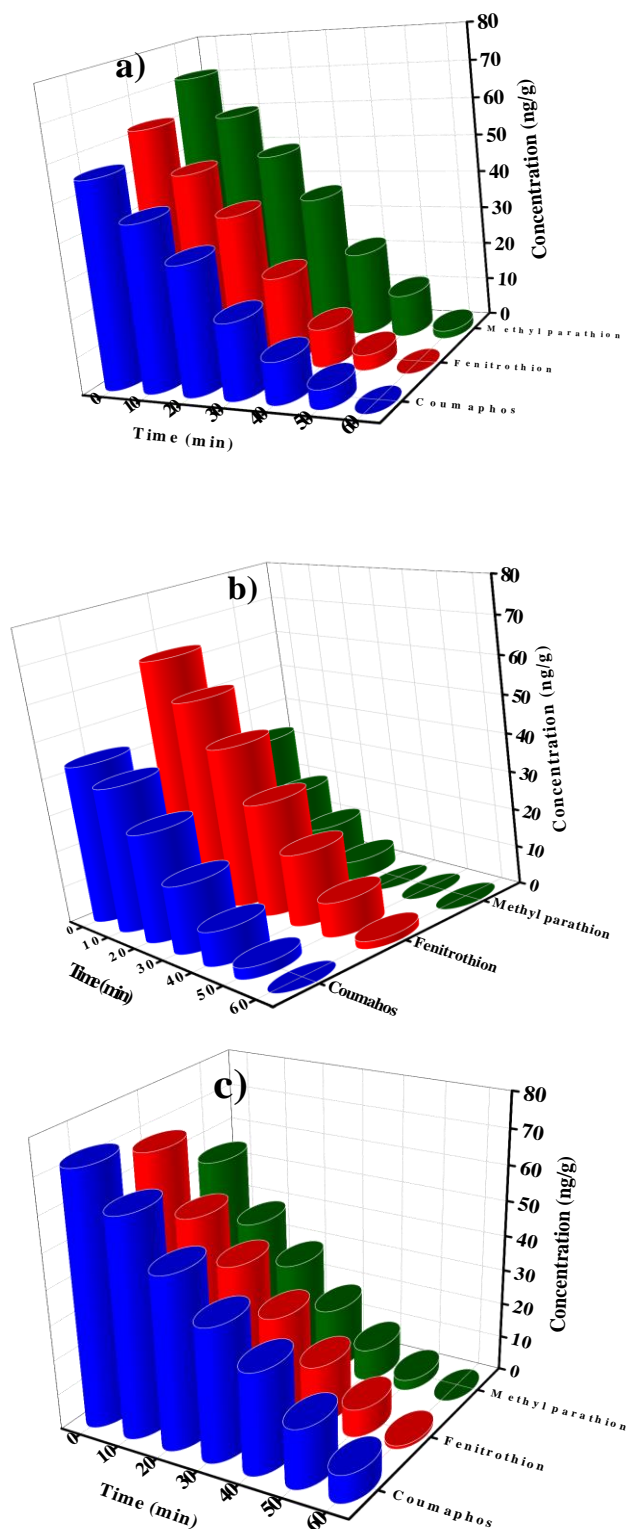
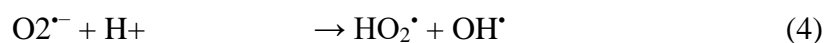
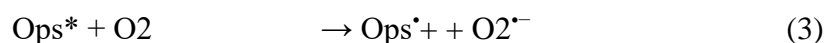


Figure 27. Photodegradation evaluation of OPs pesticides in honey under solar light irradiations
a) Beni belaid region, b) El kennar region, c) Achouat region.

Figure 27 presents the results of OPs' photodegradation of the three samples from Beni Belai, El Kannar and Achouat. For all POs, fast photodegradation was observed after 30 min for the three different regions. For Coumaphos, the removal efficiency was 63.64%, 58.14% and 47.22% at 30 min in honey from Beni belaid, El kennar and Achouat regions, respectively. The removal rates increased with the decrease of the initial concentrations. The same trends were observed for Methyl parathion and Fenitrothion. Moreover, Coumaphos showed faster degradation after the same time of sunlight irradiation. The removal of these OPs pesticides in the honey medium by photodegradation under solar light was effective; we managed to degrade almost all existing OP pesticides in less than 1 h.

The $\bullet\text{OH}$ radical is a very powerful oxidizing agent with a standard potential of +2.8 V (Akika et al. 2018), capable of oxidizing OPs into mineral end products. $\bullet\text{OH}$ played a significant role in the removal of OPs pesticides. The maximum initial concentration of OPs detected is 72 ng/g. The concentration of OPs' standards prepared was of the same order as that found in our samples to facilitate the chromatographic monitoring of analysis during photodegradation kinetics. The reaction kinetics provided information about the reaction rates and the mechanisms (Moore et al. 1981) by which the reactants were converted to final mineral products. Photoionization of OPs by electron transfer to dissolved O_2 and subsequent $\text{O}_2^{\bullet-}$ radical formation and final $\text{HO}\bullet$ yield appears to be the principal mechanism of degradation taking place. The relevant reactions causing the degradation of OPs can be expressed as follow:



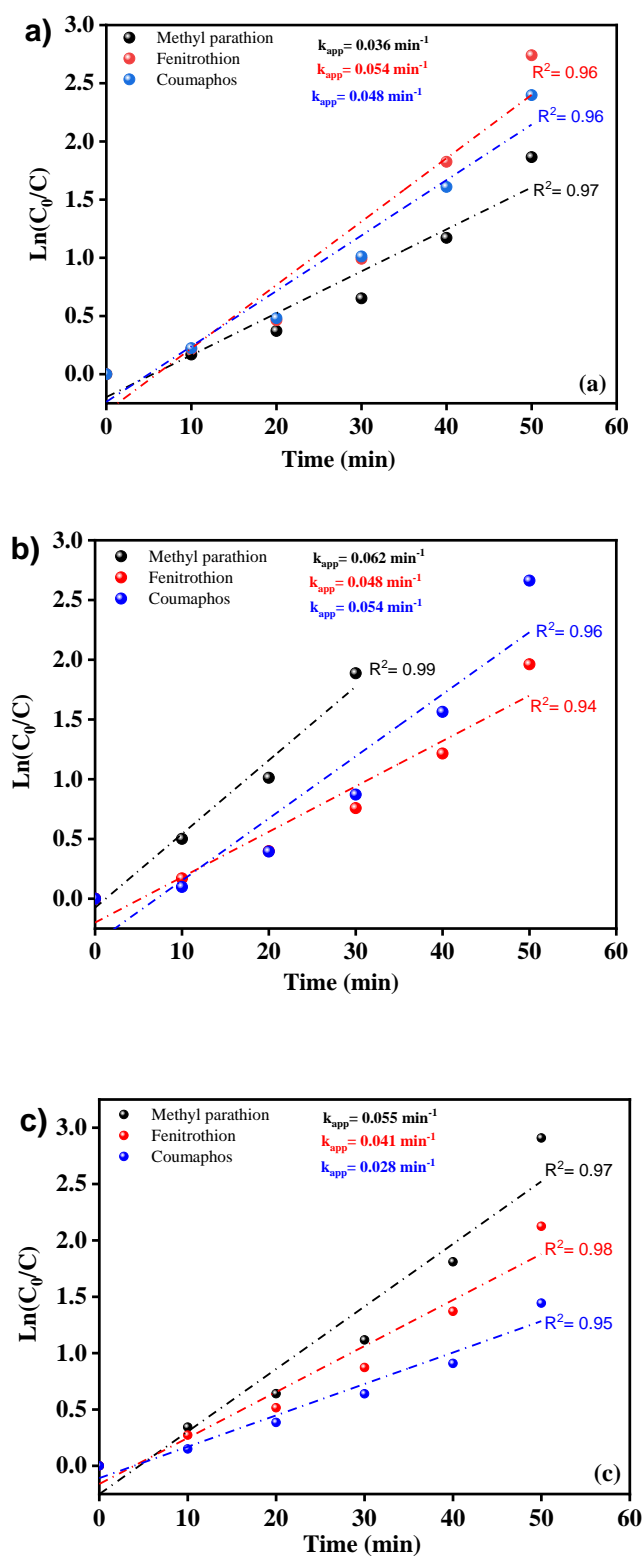


Figure 28. Pseudo-first order kinetics of photodegradation data of OP pesticides a) Beni belaid region, b) El kennar region, c) Achouat region.

The rate of photodegradation depends on the concentration; the kinetics of the photodegradation reaction of all OPs pesticides studied follows a pseudo-first order model.

$$-\frac{dC}{dt} = k_{app} C \quad (7)$$

k_{app} (min⁻¹) is the apparent rate constant, the integration of Eq. (7) gives:

$$\ln\left(\frac{C_0}{C}\right) = k_{app} t \quad (8)$$

where C_0 and C are the initial concentration and the concentration at time t of Ops, respectively.

The graph of $\ln(C_0/C)$ versus time (t) for different initial concentrations C_0 (Figure 28) corresponds well to the experimental data and, as expected, follows a pseudo-first order model. These results were consistent with previous studies where the photodegradation of organic pollutants is commonly described by first order kinetics (Yuan et al. 2014; Akika et al. 2018). The rate constant k_{app} decreased with the increase of the OPs concentration (C_0) in the honey samples studied by region, e.g., 0.062 min⁻¹ ($R^2 = 0.99$) with 33 ng/g, 0.055 min⁻¹ ($R^2 = 0.97$) with 56 ng/g, and 0.036 min⁻¹ ($R^2 = 0.97$) with 71 ng/g for methyl parathion pesticide. The highest degradation rate constant was obtained for Methyl parathion suggesting that Methyl parathion was easiest to be removed.

Conclusion and perspectives

Conclusion and perspectives

The present study was conducted to cover the following points:

- To identify the melliferous plants in the region of Jijel.
- To evaluate the quality of Jijelian honeys according to their physicochemical and bioactive properties and of Jijelian honeys and to reveal the differences between coastal and mountain honeys along with the correlation between altitude and different parameters.
- to measure the contamination of honey samples with the organophosphorus pesticides and test their photodegradation efficiency.

The investigations revealed the presence of a rich melliferous flora in Jijel region, 296 melliferous plants belonging to 69 families were identified dominated by 208 herbaceous species. These plants bloom all year round, but, April, May and June were the most abundant months with flowering melliferous plants (212, 236 and 168, respectively). 255 melliferous plants were sources of pollen and 247 were sources of nectar. In addition, the anthropogenic participation appeared in 68 planted plants participated in 23% of the total melliferous flora. On the other hand, *Galactites tomentosa* Moench, *Lotus cytisoïdes* L. and *Rubus ulmifolius* Schott were the most abundant common species in all stations.

The tested honeys had a good quality regarding physicochemical parameters, phenolic contents, and bioactive activities and they differed significantly among the samples. In addition, the antibacterial activity of 100% honey showed Jijelian honeys were efficient against *E. coli* and not a good choice against *P. aeruginosa* and *S. aureus*. The coastal samples were darker than mountainous samples and had higher pH, electrical conductivity, ash, HMF, and reducing power. Whereas, the total acidity was higher in mountainous honeys. In addition, mountain honeys did not present an antibacterial activity against *P. aeruginosa*. Hence, this study showed that coastal honeys had better bioactive potential compared to mountain honeys. Finally, the altitude presented a significant negative correlation with HMF content, electrical conductivity, ash content and pH.

The main OPs pesticides detected by gas chromatographic analysis in our collected honey samples are Coumaphos, Methyl parathion and Fenitrothion. In order to lower their environmental risk, the degradation of OPs pesticides in the honey samples was conducted in the new photoreactor using solar light irradiation. The removal of these OPs pesticides by photodegradation was

effective and existing OP pesticides were degraded in less than 1 h with a high residual percentage (> 95%). The photodegradation rate increased to 0.062, 0.054 and 0.052 min⁻¹ of Methyl parathion, Coumaphos and Fenitrothion pesticides, respectively. The kinetics of the photodegradation reaction of all OPs pesticides followed a pseudo-first order model and the relevant photodegradation reactions of OPs pesticides were reported. The sun light could be used as a source for the photodegradation method to remove OPs in the honey medium.

This research work has multiple perspectives, within these perspectives, we can cite the following:

- A larger inventory of melliferous plants is needed.
- Perform phytosociological study of melliferous plants.
- Further research on the physiochemical properties of honey is recommended and important in order to establish the criteria of assessing the quality of honey.
- A detailed study of different honey compounds is recommended (sugars, phenolics.....).
- An in vivo study is very recommended.
- Evaluate other beekeeping products (pollen, royal jelly.....)
- Check for other contaminants (heavy metals, antibiotics.....).

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Inventory of melliferous plants and physicochemical analyzes of honeys from the region of Jijel.

ملخص

منطقة جيجل هي المنطقة الأكثر خضرة في الجزائر وهي غنية بالتنوع البيولوجي والموارد الطبيعية التي تحتاج إلى تقييم وتقدير. كان الهدف من هذه الدراسة هو تقييم الإمكانات الهائلة للنباتات في جيجل وجودة عسل جيجل وفقاً لخصائصها الفيزيائية والكيميائية الحيوية والتلوث بالمبيدات إلى جانب تأثير الارتفاع على هذه القياسات. تم جرد 296 نباتاً عسلياً ينتمي إلى 69 عائلة تهيمن عليها الأعشاب ب نباتاً 208 وكان ماي هو الشهر الذي يحتوي على عدد أكبر من النباتات المزهرة (236 (80%)). بالإضافة إلى ذلك، كان للعسل المختبر جودة جيدة فيما يتعلق بالمعايير الفيزيائية والكيميائية والمحتويات الفينولية والأنشطة النشطة بيولوجياً وكانت فعالة ضد الإشريكية القولونية. إحصائياً كان للعسل الساحلي درجة حموضة، وموصلية كهربائية، ومحتوى رماد، وكثافة اللون، ومحتوى HMF، وقوة إرجاع أعلى من العينات الجبلية، بينما كانت الحموضة الكلية أعلى في عسل الجبل ($p < 0.05$). ارتبط الارتفاع بشكل سلبي مع محتوى HMF، التوصيل الكهربائي، محتوى الرماد، الأس الهيدروجيني ومبيدات الفوسفور العضوي. من ناحية أخرى، تم العثور على عينات بمستويات بقايا المبيدات الفوسفورية العضوية تتجاوز الحدود القصوى للمخلفات ≤ 50 نانوغرام/غرام التي تفرصها المياه المعيارية (منظمة الصحة العالمية). ومع ذلك، كان التحلل الضوئي بواسطة الضوء الشمسي للمبيدات الفوسفورية العضوية في العينات الملوثة فعالاً. **الكلمات المفتاحية:** جيجل، نباتات عسلية، عسل ساحلي وجبلي، خواص فيزيائية كيميائية، مضادات للأكسدة وأنشطة مضادة للبكتيريا، مبيدات الفوسفور العضوي، تحليل ضوئي.

Abstract

The region of Jijel is the greenest region in Algeria and it is rich in biodiversity and natural resources that need to be evaluated and valorized.

The aim of this study was to evaluate the melliferous potential of the flora in Jijel and the quality of Jijelian honeys according to their physicochemical and bioactive properties and pesticides contamination along with the altitude effect on these parameters.

296 melliferous plants belonging to 69 families were identified dominated by 208 herbs and May was the month that has the higher number of blooming plants (236 (80%)). In addition, the tested honeys had a good quality regarding physicochemical parameters, phenolic contents, and bioactive activities and they were efficient against *E. coli*. Coastal honeys had statistically significantly higher pH, electrical conductivity, ash content, color intensity, HMF content, and reducing power than the mountainous samples, while the total acidity was higher in the mountain honeys ($p < 0.05$). The altitude was significantly negatively correlated with HMF content, electrical conductivity, ash content, pH and organophosphorus pesticides. On the other hand, samples were found with organophosphorus pesticide residue levels exceeding the maximum residue limits (MRL ≥ 50 ng/g) imposed by the standard water (WHO). However, the photodegradation by solar light of organophosphorus pesticides in contaminated samples was efficient.

Key words: Jijel, melliferous plants, coastal and mountain honeys, physicochemical properties, antioxidant and antibacterial activities, organophosphorus pesticides, photodegradation.

Résumé

La région de Jijel est la région la plus verte d'Algérie et elle est riche en biodiversité et en ressources naturelles qui doivent être évaluées et valorisées.

Le but de cette étude était d'évaluer le potentiel mellifère de la flore de Jijel et la qualité des miels Jijéliens en fonction de leurs propriétés physicochimiques et bioactives et de la contamination par les pesticides ainsi que l'effet de l'altitude sur ces paramètres.

296 plantes mellifères appartenant à 69 familles ont été identifiées dominées par 208 herbes et mai a été le mois qui a le plus grand nombre de plantes en fleurs (236 (80%)). De plus, les miels testés étaient de bonne qualité en ce qui concerne les paramètres physicochimiques, le contenu phénolique et les activités bioactives et ils étaient efficaces contre *E. coli*. Statistiquement, les miels côtiers avaient un pH, une conductivité électrique, une teneur en cendres, une intensité de couleur, une teneur en HMF et un pouvoir réducteur significativement plus élevé que les échantillons montagneux, tandis que l'acidité totale était plus élevée dans les miels de montagne ($p < 0,05$). L'altitude était significativement négativement corrélée avec la teneur en HMF, la conductivité électrique, la teneur en cendres, le pH et les pesticides organophosphorés. En revanche, des échantillons ont été trouvés avec des niveaux de résidus de pesticides organophosphorés dépassant les limites maximales de résidus (LMR ≥ 50 ng/g) imposées par l'eau standard (OMS). Cependant, la photodégradation par la lumière solaire des pesticides organophosphorés dans les échantillons contaminés était efficace.

Mots clés: Jijel, plantes mellifères, miels côtiers et montagnards, propriétés physicochimiques, activités antioxydantes et antibactériennes, pesticides organophosphorés, photodégradation.