

Etude Physico-chimiques et Analyses Chromatographiques de l'huile Essentiel des Grains de céleri (*Apium Graveolens.L*) [Physico-chemical and Chromatographic Analysis Study of the Essential oil of Celery Seed (*Apium Graveolens.L*)]

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Abstract: *The study of oils is carried out on the species (*Apium graveolens. L*) in the area of Marrakech. We started with the extraction of essential oil by the method of hydro-distillation. The physicochemical results of the characterization for the essential celery oil answer all the limits recommended by the standards of AFNOR. We particularly quote the acid value, the index of ester, saponification, peroxide, iodine and the index of refraction. Also we analyzed our oil by gas chromatography whereas we found a product majority that of D-Limonene with 79.15%.*

Keywords: *Celery graveolens.L, Oil essential, hydro-distillation.*

1. INTRODUCTION

Celery (*Apium graveolens.L*) Is a plant of the family of Apiécées or Umbellifers. This family also comprises carrot, fennel, dill, parsley, anise, cumin, etc the three shapes of celery are usually cultivated: celery-connects or ribbed celery, the celeriac, celery to be crossed or small celery. This vegetable can be found in food in various forms (such as it is, out of powder, celery seeds, salt to celery...) [1].

Ingestion of celery frequently leads to intolerance reactions in inan, a fact that has been known for almost 70 years [1-9]. The symptoms comprise local reactions (oral itching, inflammation of the lips and oral mucosa, angioedema of the throat, gastroenteritis, and diarrhoea), and a considerable number of patients experience systemic symptoms (urticaria, asthma, anaphylactic shock).

Celery contains vitamin C and other materials which are health substances improving such as phthalide, which lowers cholesterol and coumarins which help to prevent cancer. Celery seeds of anti-rheumatism, sedative, disinfectant of the urinary tracts, increase in the excretion of the uric acid, the pressure blood to drop, to a certain extent against the fungic diseases, diuretic, analgesic, anti-inflammatory drug, the detoxication, antispasmodic anti-bacteria, of the rules constraining, anti-contractions, of the convulsions, tonics of the stomach and carminative. [10]

In this work, we propose to analyze by gas chromatography, in order to see conformity according to the standards recommended by the pharmacopeia, of an essential celery oil, intended for a therapeutic use.

In a first part, the oretical outline on essential oils by gas chromatography (CPG), are given.

After a familiarization with a technique of analysis impossible to circumvent in fact gas chromatography CPG. We carried out the analysis of the essential celery oil. Thus calculate for the physicochemical characteristics by proportioning acid-bases.

2. EXPERIMENTAL PART

2.1. Vegetable Equipment

Equipment vegetable is made up by the grains wall and dry Celery graveolens. L. the fruits walls collected in the area of Marrakech in a movable, rich and wet ground in the following part are dried with the sun during approximately a week. Thus are dried with the drying oven at the weighed temperature of 40°C hanging two days then, is then ready for the extraction.

2.2. Extraction of Oil

The system of hydro distillation clevenger is the equipment used during extraction of celery seeds (Apium graveolens.L). The steam which assembles balloon condenses in cool and changes in the form of droplets (water + oil) in one the second time oil will be recovered at the end by a decantation by elimination of water and obtaining desired oil the extraction made continuously is of approximately three good fortunes.

2.3. Analysis Physicochemical

Determination of the acid value: (NF ISO 1242: 1999 (T 75-103)) [11] It is the number of Mg of KOH necessary to the neutralization of the free acids contained in HE 1g. The free acids are neutralized by EtOH a titrated solution of KOH.

Determination of the ester index: (AFNOR NF T 75 - 104: 1994) [11] It is the number of Mg of KOH necessary to the neutralization of the acids released by the hydrolysis of esters contained in HE 1g. The hydrolysis of esters present in HE is done by heating, under defined conditions, in the presence of a EtOH solution titrated of KOH and back titration of the alkali excess by a graduated solution of HCl.

Determination of the index of refraction: (AFNOR NF ISO 280: 1999 (75-112)) [11] It is the relationship between the site of the angles of incidence and refraction of a light ray determined wavelength, passing from the air in HE maintained at a constant temperature.

Determination of the index of saponification (NF ISO 3657: 1999 (T57-105) It number of Mg of KOH necessary to saponify 1g fat. The index of saponification will be all the

Determination of the peroxide index. In the presence of oxygen in air, the unsaturated fatty-acids using the composition of the fats oxidize by giving peroxides. This phenomenon takes place during the storage of the fats: it is rancidity. The determination of the quantity of peroxides of a fat shows its deterioration by oxidation. The peroxide index is defined as being the number of milliéquivalent of oxygen in kilograms of fat.

Determination of the iodine index of a fat by the iodometric technique, it is the number of gram of iodine which reacts with 100g of grease.

2.4. Analysis Chromatographic

Operating conditions of Gas chromatography (CPG): The chromatographic analysis of HE was carried out with a chromatograph in gas phase type chromatography in gas phase 7890b - agilent technologies the fragmentation is carried out by electronic impact with 70eV. The column used is a capillary tube HP 5MS (30mx0, 25mm). The thickness of the film is of 0,25µm. The temperature of the column is programmed of 50 with 250°C at a rate of 4°C.min-1. The carrier gas is the helium whose flow is fixed at 1.5 ml.min-1. The mode of injection is mode Split (report of escape: 1/70). The device is connected to a computer system managing a library of spectrum of mass NIST 98 and is controlled by a software "HP ChemStation" making it possible to follow the evolution of the chromatographic analyses. The identification of the components was made on the basis of comparison between their indices of retention and those of the standard compounds of the computerized data bank (NIST 98). [12]

3. RESULTS AND DISCUSSION

The output of extraction varies from 0.52 to 0.9% starting from the grains by the method of hydro-distillation; two independent factors could affect the output of extraction: quality of the raw materials, and the procedure of extraction. It was observed that the crushing of the grains increased the output of

extraction. It is known that essential oil is hydrophobic, whereas the substances are soluble in liquid carbon dioxide, consequently, it can be provided that part of oil will be isolated starting from the vegetable matter.

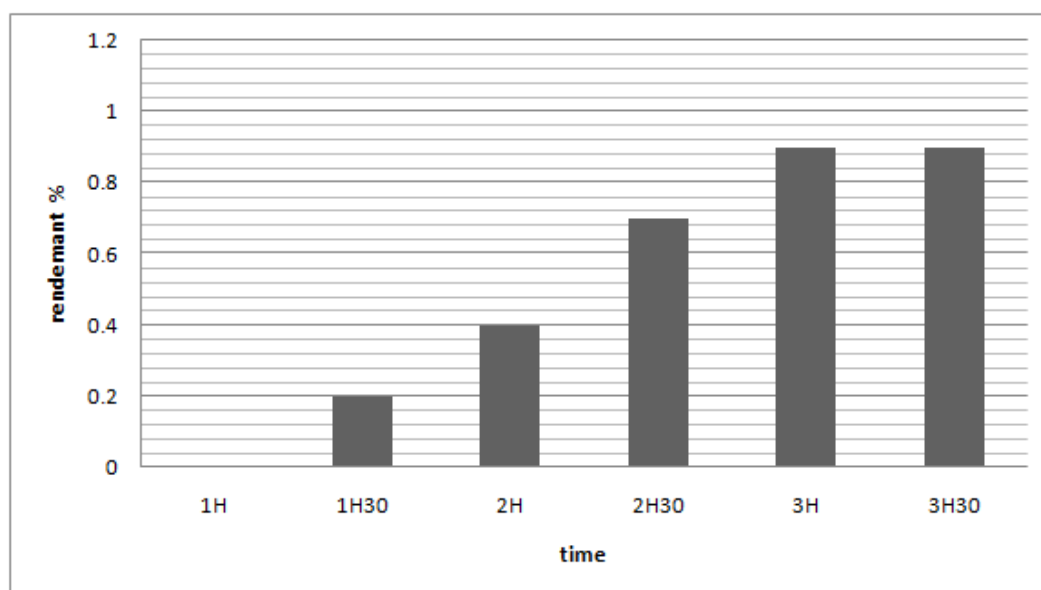


Figure1. Extraction of the celery oil according to time

3.1. Physico-chemical Characteristics

From these values, this reveals that all these constants being influenced by the edaphic and climatic conditions as well as the cultivation methods [13] [14] [15]. That been part of the complexity of the concept of chémo-type.

However and in spite of these fluctuations, we notice that the physicochemical parameters of our HE are in agreement with those mentioned by the standards.

For the chemical constants, the acid value gives an idea on the free rate of acids. In our study, this index, certainly in the standards, remains relatively high. That can find an explanation in the degradation of HE (hydrolysis of esters) during its conservation, which is prejudicial in the long term. Conversely, a IA lower than 10 is a proof of good conservation of the petrol (small quantity of free acids). [16]

An index of refraction varying primarily with the content of monoterpenes and oxygenated derivatives. A strong content monoterpenes will give a high index. For certain authors [16], the weak index of refraction of HE indicates its weak refraction of the light what could support its use in the cosmetic products.

The determination of the physicochemical properties is a stage necessary but remains nonsufficient to characterize HE. It will be thus paramount to determine the chromatographic profile of the aromatic petrol.

Table1. physicochemical indices for *Apium graveolens.L* oil

index	Test N°1	Test N°2	Test N°3	Average
Acid index	6,47mg/g	6,59mg/g	6,12mg/g	6,39mg/g
Peroxyde index	10 meq.O ₂ /kg	8,86meq.O ₂ /kg	9,87meq.O ₂ /kg	9,57meq.O ₂ /kg
Saponification index	72,54 mg/g	79,80 mg/g	68,29 mg/g	73,54 mg/g
Iodine index	20,6 mg/g	19,72 mg/g	20,18mg/g	20,16 mg/g
réfractive index	1,31466	1,31430	1,31489	1,31461

For the acid value (table 1) shows fatty acid in oil. A value is very high is prejudicial with edible oils, which shows that it does not have sudden a degradation. And for the index of saponification it indicates to us that it is less rich in fatty-acid length of average chain (acid steric with 18C). For the iodine index it has a very small quantity of fatty acid with an average number of non-saturation. To the end the results for the peroxide index indicates that it does not have sudden deterioration.

3.2. Chromatographic Analysis

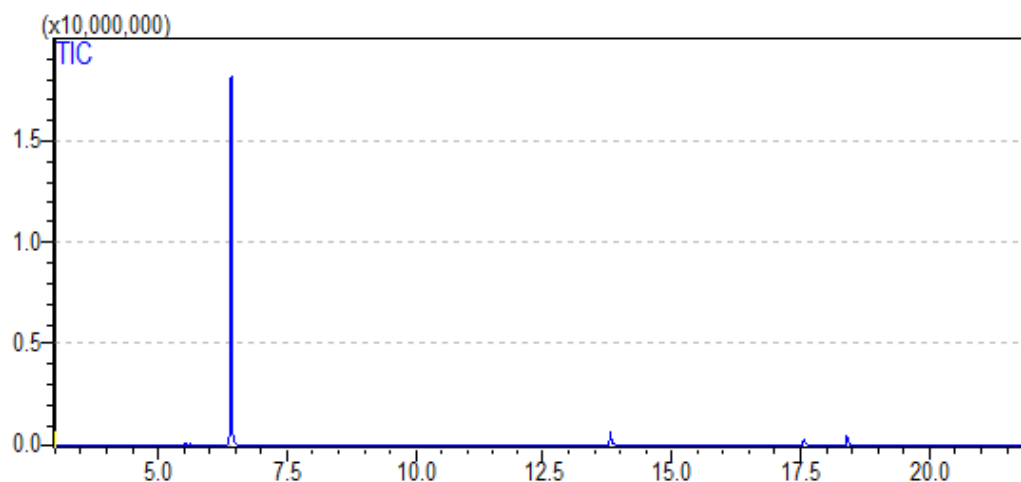


Figure2. chromatogram for oil of the Celery graveolens grains. L

The results of the analysis by Gas chromatography of the chemical composition of essential oil are presented in the table 2, in which the identified compounds are listed according to the order of prevalence. On the whole, 12 compounds were identified what corresponds to a percentage of 94,99% compared to the whole of the component isolated. D-Limonene seems the constituent majority one of the HE (79.15%), followed by Alpha-Hydroxypropylbenzene (4.23%), of the β -Selinene (9.25%), of Bèta-Myrcene (1.93%). Speaking Biochimiquement, this HE is primarily made up of a mono-terpene (80%). Sesquiterpenes (4%). The ketones are arather important rate (2%) made up only of 2-chloro-1-(-2,4-dimethylphenyl) - 2-methyl-1-propanone. Lastly, the minority compounds belong to the family of oxides, aldehydes and alcohols sesquiterpenic. (Table 3)

Table2. Biochemical classes of the compounds identified in HE

Identification	Type of molecule	Chemical formula
Alpha-Thujene	Monoterpène	C10H16
Beta-Pinene	Monoterpène	C10H16
Bèta-Myrcene	Monoterpène	C10H16
D-Limonène	Monoterpène	C10H16
β -Selinene	sesquiterpène	C15H24
Caryophillene oxide	sesquiterpène	C15H24

Table3. Composition (%) of the volatile fraction of the extracts of celery grains

identification	%
Alpha-Thujene	0,48
Hydroxybutric acid lactone	0,46
Beta-Pinene	1,04
Bèta-Myrcene	1,93
D-Limonène	79,15
6-butyl-1,4-cycloheptadiène	0,64
Amyl benzene	0,96
β -Selinene	3,73
Caryophillene oxide	0,28
Beta-Selineol	0,33
2-chloro-1-(-2,4-dimethylphenyl)-2-methyl-1-propanone	1,76
Alpha-Hydroxypropylbenzene	4,23
Total Percentages	95%

4. CONCLUSION

The nonconventional oils extracted seeds wall celery (Celery graveolens. L), collected in the area of Marrakech. This oil resembles itself by their physicochemical properties and their compositions by the method chromatographic.

The oil of color yellow clearly, liquid with room temperature, present of the physicochemical characteristics seems with those measured by the international standards according to AFNOR. This oil is of unsaturated type and contains mainly the oleic fatty-acids C18 and palmitic acids C16.

Taking into account these results, the Celery *graveolens*. L can be cultivated for the production of technical oil of interest (drug, insecticides, perfume, fungicide).

The results got not CPG show that analyzed oil contains several compose. We identified four elements whose times of retention are identical to those found in analyzed essential oil.

They correspond to the principal chemical compounds of the essential oil of celery found in the literature of which only one intense peak corresponding to D-Limonene.

REFERENCES

- [1] Heidi Collombier, "La cuisine Sauvage" [archive], 2008-2010 (accessed January 13, 2010): "Tender and aromatic, the young leaves of egopod make very good salads, tasty vegetables cooked Steam or succulents soufflés."
- [2] Jadassohn, W. & Zaruski. M. (1926) Idiosyncrasie gegen Sellerie, Arch. Dermatol. Sjph. 151, 93-97.
- [3] Kretzner, M. & Lindemayer, J. W. (1983) Sellerieallergie (Selleriekontakturtikaria Syndrom) und Zusammenhänge mit Allergien gegen Pflanzenallergene, Wiener Klin. Wschr. 23, 838- 843.
- [4] Wuethrich. B. & Hofer, T. (1 984) Das „Sellerie-Beifuss-GewurzSyndrom". Dtsch. med. Wschr: 25, 981 -986.
- [5] Pauli, G., Bessot, J. C., Dietermann-Molard, A., Braun, P. A. & Thierry, R. (1 985) Celery sensitivity: clinical and immunological correlations with pollen allergy, Clin. Allergy 15, 273-279.
- [6] Vallier, P., Dechamp, C., Vial, O. & Deviller, P. (1988) A study of allergens in celery with cross-sensitivity to mugwort and birch pollens, Clin. Allergy 18, 491 -500.
- [7] Wuethrich, B., Staeger, J. & Johansson, S. G. O. (1990) Celery allergy associated with birch and mugwort pollinosis, Allergy 45, 566-571.
- [8] Staeger, J., Wuethrich, B. & Johansson, S. G. O. (1991) Spice allergy in celery sensitive patients, Allergy 46, 475-478.
- [9] Pauli, G., de Blay, F., Bessort, J. C. & Dietermann, A. (1992) The association between respiratory allergies and food hypersensitivities, Allergy Clin. Immunol. Neivs 4, 43-47.
- [10] Ebner, C., Hirschwehr. R., Bauer, L., Breiteneder, H., Valenta, R., Ebner, H., Kraft, D. & Scheiner, O. (1995) Identification of allergens in fruits and vegetables. IgE-crossreactivities with important birch pollen allergens Bet v 1 and Bet v 2 (birch profilin), J. Allergy Clin. Immunol. 95, 962-969.
- [11] Aramaini D. Reduced serum testosterone in healthy women, steroids. 2003,69: 763-770.
- [12] AFNOR. "Compendium of standards: essential oils. Volume 2. Monographs on essential oils. AFNOR, Paris, 2000, 661-663.
- [13] DEMARNE FE. « Le géranium rosat ». Parfums, Cosmétiques et Arômes, n°62, 1985.
- [14] GILLY G. « Les plantes à parfum et huiles essentielles à Grasse ».
- [15] LIS-BALCHIN M. « Geranium and pelargonium: the genera *Geranium* and *Pelargonium* ». CRC Press, Taylor & Francis, London, 2002, pp: 116-131, 147-165, 184-217.
- [16] KANKO C, SAWALIHO BE, KONE S, KOUKOUA G, N'GUESSAN YT. « Étude des propriétés physico-chimiques des huiles essentielles de *Lippia multiflora*, *Cymbopogon citratus*, *Cymbopogon nardus*, *Cymbopogon giganteus* ». Comptes rendus Chimie 7 (2004) 1039–1042.