Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2015, 7(8):337-343

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Chemical characteristics of the cladodes juice of *Opuntia ficus indica* harvested Zerhoun (Morocco) and its effect on the growth of lactic bacteria

Mohamed Bou-idra[2], Abdelaziz Ed-dra[1], Fouzia Rhazi Filali[1] and Amar Bentayeb[2]

[1]Team: Microbiology and Health, Department of Biology, Laboratory of Chemistry Biology applied to the Environment, University Moulay Ismail Faculty of Sciences, Zitoune Meknes, Morocco
[2]Team: Physico-chemistry of Condensed Matter, Faculty of Sciences, University Moulay Ismail, Meknes, Zitoune Meknes, Morocco

---

ABSTRACT

The juice of the cladodes of Opuntia ficus indica whose composition highlights an interesting content in secondary metabolites, fibers and mineral substances, can justify its use for the proliferation of lactic acid bacteria during fermentation. The kinetics of growth of these two bacteria : Streptococcus sp and Lactobacillus sp shows that in the presence of juice of cladodes of Opuntia ficus indica to 5 %, the values of the absorbance at 600nm peaks are present much more important than those of the control sample in its absence. The statistical analysis through the software Excel 2007 results, confirmed that there is a very signify difference between the growth of control sample and the one with the presence of 5% of the juice of cladodes of Opuntia ficus indica, either p=0.00356991 for the case of Lactobacillus sp and p= 0.008129737 for Streptococcus sp.

Keywords: Opuntia *ficus indica*, juice cladodes, phytochemistry, lactic bacteria.

INTRODUCTION

The Opuntia cactus is a plant native to Mexico [1]. It was introduce to the south of Spain following the first shipment of Columbus to the New World. Subsequently, it has been spreading throughout the Mediterranean Basin during the 17th century [2]. With the exception of the Saharan areas and mountainous, the Opuntia are widely represented in the rural landscape of Morocco, in addition to less regular plantations, and around villages or in plants of closure limiting the plots of crops or orchards [3]. Culture of Opuntia exists in almost all regions of the country with the areas relatively variable, it occupies an area estimated at 54530 ha and represents 11.07% of the total area under fruit trees [4]. Its geographical distribution is broad enough since it is found in coastal areas from Sidi Ifni up to Tangier, that in several continental regions [4]. The best plantations are located in coastal areas, and more specifically in the coastal strip of more than 10km of with that undergo the maritime influence. The plant is in fact hardy to the fog and night morning, very common in this area, throughout the year [4].

The *Opuntia ficus indica* or prickly pear is a plant of type CAM (Crassulacean Acid Metabolism) [5], who presented adaptations of physiological and morphological, allowing him to withstand the difficult conditions of the arid and semi-arid lands.

It is currently also integrated in the strategies for combating desertification in different countries [6; 7; 8; 9 and 10].

In some countries, the *Opuntia ficus indica* is the subject of a culture in its own right, for the exploitation fodder and vegetable [11 and 12]. In effect, it is a plant that all parties can be consumer: the rods (or cladodes) and the fruit.

The smooth variety submitted by report to the variety thorny, has the advantage of being easily maniple and exploited as fodder. The cladodes may constitute 35% to 40% of the food ration for basis [13] and can constitute a source of water for the animals. In the case of lambs fed with a large share of *Opuntia*, the water needs are virtually zero [9]. They can also be use in the feeding of small animals [14].

Thus, this plant is a potential candidate for the development of functional foods and healthy foods [15; 16], replying to the request of the consumer, in balanced food in calories, cholesterol, fat, dietary fiber and antioxidant.

In order to take advantage of the potential nutritional and medicinal potentials of cladodes of *Opuntia*, their incorporation in juice form in the food, could be an alternative for their exploitation [17]. In effect, the juice of nopal has a high content of pectin and mucilage, these substances are transformed into gel once ingested, and will allow the fats to transit through the digestive system without being absorbed. In other words, the juice of nopal has a good thinness effect, thanks to its effect gelling agent in the gastrointestinal system; the juice of nopal is able to reduce the level of sugar in the blood. It is the ideal base for dietary supplements controlling the blood glucose levels of diabetes type II.

Taking into account the surprising nutritional qualities from this plant, its hypoglycemic and lipolytic characteristics very sought after in the natural food additives, we anticipate introducing this juice as an ingredient in dairy products rich in fat and sugar, to make them more digestible and more adapted to the malfunction of our energy metabolism.

Our work in this perspective; we have addressed the impact associated with the addition of the juice of cladodes, in the bacterial culture medium, on the improvement of the growth of lactic acid bacteria, in order to consider studies to optimize the consistency of the clot, to enrich the nutritional characteristics of dairy products and to improve their taste quality.

EXPERIMENTAL SECTION

Sampling Site of the biological material

The cladodes of Opuntia taken at the foothills of the Bouassal Zerhoun area located North West of Morocco. This plant covers a geographical area about 636 km^2. The region enjoys a Mediterranean climate, semiarid, suffering continental influences, resulting in high thermal amplitude. The highest average temperatures between spring and autumn range from 14°C to 20°C, while the average minimum temperatures range from 5°C to 14°C. The average maximum temperatures in summer rise to 42°C. Winter is relatively cold in December, January and February; the minimum and maximum average temperatures are respectively 2^oC and 11°C [18]

The removal of cladodes conducted during the months of March to May the years 2014-2015, for 10 shrubs. These cladodes aged 1 to 10 months, are classified in 3 batches whose weight are between 100g and 400g. The samples are stored at 4°C for a period not exceeding 3 days before their analysis and exploitation. This selection allows the choice of stages whose composition satisfies the maximum nutraceuticals needs [17]

Preparation of the plant material

Extraction of juice

The cladodes cleaned, trimmed and their juice extracted and separated by centrifuge (Brand Robotic) at 6200g. The juice collected represents 30% to 38% of the fresh weight (W/W). It filtered on Whatman paper N°.4, to collect the juice, and then sterilized by filtration through a pecto-cellulosic membrane of porosity 0.45 µm before being stored at -18 °C.

Phytochemical study

The phytochemical tests have been performed on different extracts prepared from cladodes *of Opunita ficus indica* by using specific reagents of revelation.

The screening phyto has allowed us to highlight the presence of secondary metabolites at the level of plant tissues of this plant.

The detection of these chemical compounds based on tests of solubility of constituents, reactions of precipitation, turbidity and a change of specific color [19- 25]

Chemical analysis

The dry matter (DM) content was determined at a temperature of 105°C [26] and the content of inorganic materials by incineration at 550°C for 4 hours [27]. The concentration of Brix (soluble solids) was determined by refractometer (brix RHB-32QTC) at 20°C.

The acidity was measured according to the standard method (NFV05-101- 1974) with 0.1N NaOH to pH 8. The results are expressed as % cladodes juice.

Bacteriological analysis

Test of the antibacterial effect of juice of the cladodes

The technique of Hayes and Markovic (2002) [28], is to remove a sterile disk, soaked in juice of cladode undiluted, on a microbial mat at the very beginning of its growth and measure the area where the microorganisms have not been able to develop. The diameter of inhibition, which translated the antimicrobial activity of juice, is thus determined and compared to the standard values dictated by the standards.

A microbial suspension (of different pathogenic bacteria belonging to the Gram+ and Gram-) of equivalent density standard of 0.5 Mac Farland (10^8 CFU /ml) is prepared as preculture. 20 ml of agar (MHA) are be sunk by Petri dish. After solidification, two ml of inoculum were deposited on each of them. After impregnation of 5 minutes, the surplus of inoculum was removed by suction. On each box, a sterile disc of filter paper of 6 mm diameter, soaked in 20 µl of juice was deposited on the surface.

The Petri dishes lifted 1 hour at ambient temperature, and then incubated at 37°C for 18 to 24 hours. The diameter of inhibition is measured in millimeters, disk included. The number of repetition is three times.

Effect of the juice on the growth of lactic bacteria

This study is to monitor the behavior of lactic bacteria *Lactobacillus sp* and *Streptoqoccus* sp, isolated and purified separately from a yogurt commercial, in the presence of increasing concentrations (V/V) of juice of cladodes, either respectively 0 %, 1 %, 2.5 %, and 5% in relation to the volume of culture medium (Nutrient broth, Biockard).

The inoculation is done by introducing 0.1 ml of preculture of lactic bacteria (*Lactobacillus sp* or *Streptoqoccus sp*), which is in exponential phase of bacterial growth and with density is 10^7CFU/ml, in 100 ml of nutrient broth. The mixture was incubated at 37°C for *Lactobacillus sp* [29 and 30] and to 42°C for *Streptoqoccus sp* [31]. The bacterial growth evaluated by following the values of the optical density of the bacterial culture at 600nm as a function of time, the processing of data and statistical analysis performed with the software Microsoft Office Excel (2007).

RESULTS AND DISCUSSION

Phytochemical Screening

The results of the phytochemical characterization tests on the studied prickly pear cladodes reveal the presence of leucoanthocyanins and various compounds such as C-heterosides, mucilage, monosaccharides and holosides, sterols, triterpenes and heteroside cardiotonice, but we noted the total lack alkaloids, tannins, saponins, anthocyanins, free and combined anthraquinones and drugs (Table 1). These results are similar to the work done by Dib and al, (2013) on *Opuntia ficus indica* Algerian, who have also noticed the absence of Saponoside, anthracyanosides, anthracenosides and coumarins, but in contrast, they noted the presence of alkaloids and tannins contrary to what we found in this study. This may suggest that the composition of the cladodes of Opuntia can change depending on the geographical distribution and the climatic conditions.

The presence of flavonoids in the cladodes constitute, by the number of compounds and the quantities present, the main class of polyphenols anti-oxidants and sensors of free radicals. They are particularly abundant in foods and beverages such as grapes and the preparations derived, tea and cocoa , which are deemed to have protective effects against various chronic diseases such as cardiovascular diseases , type II diabetes, cancers, neurodegenerative diseases [32]. The pharmacological properties of polyphenols in general and the flavonoids in particulars make them as popular dietary adjuvant molecules [33].

Chemical Characters of cladodes

In analyzing the results of Figure1a and 1b, we find a relationship quite logical between the weight and the size of the different lots studied. Table 2 shows a highly significant correlation between the growth parameters of the cladodes (weight, length and width) and their constituent components.

The dry matter content of the cladodes declined in the course of their growth, it evolved from 8.7% in the lot 1 to 7, 9 % in the lot 3, indicating a high water content among the latter (Table 2), which is in concordance with the results of Sadok in 2008 [17]. By comparing the young cladodes of the stage 1 (lots 1), with other plant products, we note that it is similar to the one of vegetables deciduous trees such as the chicory, spinach and celery [34].

Table 1: Results of phytochemical examination of prickly pear cladodes

The families			The organes
			The cladodes
The alkaloids	Mayer		_ _ _
	Dragendorff's reagent		_ _ _
Polyphenols	The tanins	The tanins fastness	_ _ _
		The tanins catechiques	_ _ _
	The flavonoides	Anthocyanins	_ _ _
		React. Cyanidine	_ _ _
		Leucoanthocyanes	+ + +
Derivatives anthraceniques free			_ _ _
Derivatives anthraceniques combined		The O-glycosides	_ _ _
		The C-glycosides	+ + +
Sterols and triterpenes			+++
Mucilages			+ + +
Saponosides			_ _ _
Glycosides cardiotonics			+ + +
The compounds reducers			_ _ _
Raunchy and holosides			+ + +

+++: Strongly present; ++: Moderately present; (+) : weakly present ; (-): negative test

The percentage in mineral substances increased significantly during growth of the cladodes, either respectively of 15.3% in the first batch to 16.55% in the third. According to the literature, it is higher than that of vegetables of common use [35]. These results are similar to the work done on *Opuntia ficus indica* Algerian by Sadok (2008) and Nezfaoui and Chermiti (1991), [36 and 17]. These authors have also reported that the mineral is important in the cladodes of less than a year; it varies so highly significant from 12.00 to 15.49% in relation to the dry matter. This shows the richness of the cladodes of the zone of Zerhoun area of Morocco in which mineral atlantes' 16.55 %.

Concerning the soluble dry extract of the juice of cladode, the results obtained (Table 2) show that it decreased during growth of 2.53% in batch 1 to 2.04 % in the 3rd batch. This content in soluble dry extract lowed compared to that of fruit pulp that is 12% - to 17 % [17]. However the work achieved by Nefzaoui and Chermiti (1991) [36 and 17] have shown that the cellulose content (fiber) increased significantly during the stages of growth from 8.84 to 11.62 g/100 g of dry matter, and are higher than those of lettuce and spinach which contain less than 3% of the dry matter.

For acidity, it presents very little significant variation in the course of the evolution of the growth of the cladodes, which can have a negligible influence on the lactic fermentation.

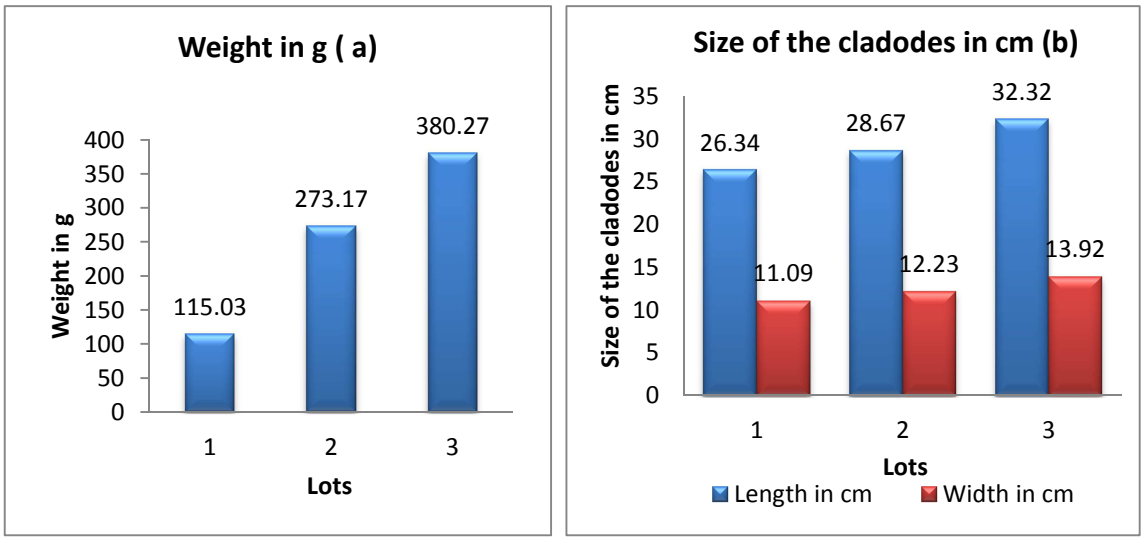

Figure 1: Variation of the size and weight of the cladodes in function of the age represented by batches

Table 2: Results of chemical examination of prickly pear cladodes

Lots	1	2	3
Acidity in %	0.97 ± 0.01	0 .93 ± 0.02	0.98 ± 0,011
Soluble dry extract in% (Brix)	2.53 ± 0.01	2.28 ± 0.02	2.04 ± 0,015
Dry Matter in %	8.7± 0.1	8.16 ± 0.16	7.9 ± 0.12
Mineral Matters in %	15.3 ± 0.02	16.02 ± 0.03	16.55 ± 0.05

Microbiological testing

Antibacterial effect of juice of cladodes

The study of the antibacterial effect of juice *of Opintia ficus indica*, has shown the absence of the antibacterial activity on the 6 strains of pathogenic bacteria belonging to both Gram-positive that the Gram-negative: (*Pseudomonas aeruginosa*, *E. coli*, *Streptococcus feacalis*, *Klebsiella pneumonae*, *Staphylococcus aureus*, *Salmonella spp)* Table 3.

Table 3: Results of the antibacterial testing of prickly pear cladodes juice: (-): absence of zone of inhibition growth. (+): Presence of zone of inhibition growth, measured in mm

	Tries 1	Tries 2	Tries 3
Pseudomonas aeruginosa	-	-	-
E. coli	-	-	-
Streptococcus feacalis	-	-	-
Klebsiella pneumonae	-	-	-
Staphylococcus aureus	-	-	-
Salmonella spp	-	-	-

The cladodes juice effect on lactic acid bacteria (*Streptococcus sp* and *Lactobacillus sp*)

We find that the addition of the juice of cladode to bacterial cultures of *Streptococcus* sp and *Lactobacillus sp* is in favor of an increase in the speed and bacterial cell density. This increase of bacterial growth is a function of the concentration of juice tested, in the culture medium (Figure 2). The statistical analysis of the results, has shown that there is a significant difference between the speed of bacterial growth of the control samples, and the one carried out in the presence of juice of cladodes of *Opuntia ficus indica* to 5%, representing a value of p=0.0035 for the case of *Lactobacillus and* p=0.0081 for the *Streptococcus*. These results are comparable to those found by Sadok (2014) [35] in a study prepared in Algeria, concerning the effect of the juice of cladodes on the fermentation of milk.

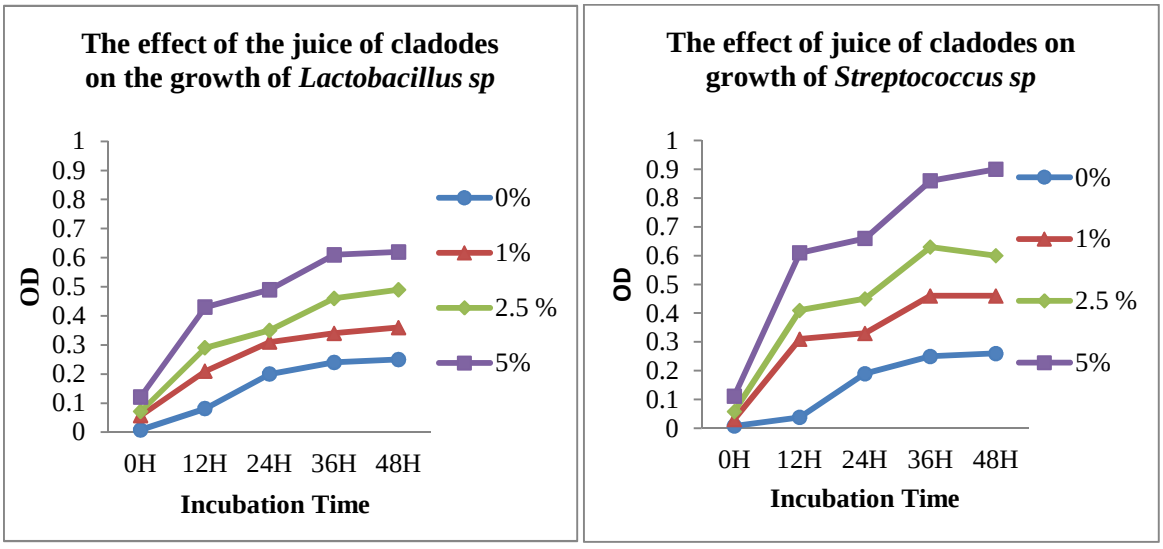

Figure 2: Graphical Representation of the evolution of the growth of *Streptococcus sp* and *Lactobacillus sp, expressed as* OD at 600nm, as a function of the concentration of the juice of the cladodes *of Opuntia ficus indica*

Based on the results of the chemical composition of the cladodes of Opuntia harvested from the area of Zerhoun, and that of the literature, we find that this part of the plant much enriched in components with a beneficial effect on health.

The mucilage contains of polygalacturonic acids that are similar to pectin playing an important role in the contribution of dietary fiber and in the balance of the intestinal transit, by stimulation of the growth of beneficial

bacteria in the colon (probiotics) [37]. The mucilage contains neutral sugars such as the D-xylose, lactose, the rhamnose, L-arabinose [38], which may be a nutritional alternative in the case of an energy metabolism failure. The leucoanthocianes are pigments that contain active principles to maintain and preserve the vascular system. [39]. The cladodes were also used for their medicinal effect as anti-inflammatory [40 and 41], glucose and cholesterol lowering [42 and 43], antioxidant [44] and as an emulsifier in food products, pharmaceutical products and cosmetics ... [45 and 46].

Therefore, the wealth of the juice of cladodes of *Opuntia ficus indica* in nutrients, such as carbohydrates, proteins, lipids ... [31], and in mineral, vitamins such as vitamin C and dietary fiber, promote growth, and improve the proliferation of lactic bacteria [32]. It appears that the use of the juice in the dairy industry may have a beneficial effect in the consistency of the clot and the organoleptic quality and sensory data derived from milk.

CONCLUSION

The juice of the cladodes contained composition important in glycosides in holoside and raunchy, in leucoanthocianes, fibers and minerals justifying its interest.

The kinetics of the growth shows that in the presence of juice of cladodes of *Opuntia ficus indica* to 5%, the *Streptococcus sp and the Lactobacillus sp* reach an important optical density and a speed of more rapid growth, confirmed by statistical analysis of the results.

At the end of this study, it seems interesting to us to project our research on the development and optimization of lactic fermentation, to produce less expensive milk derivatives whose organoleptic and sensory quality are more attractive.

REFERENCES

[1] Mulas M, OF Hallewin G, and Canu D, **1992**. Osseervazioni sulla radicazione di cladodi di a annodi *Opuntia ficus indica* Mill." *Rivista di frutticoltura* 10: 67-60.
[2] Ayouz M, Alary V, Mekersi S, **2012**. *Agriculture* 21, 438-447.
[3] Arba M, EL Aich A, Sarti B, Belbahri L, Boubkraoui HAS, Ait Hammou Rahali, Prisoner A, et al, **2000**. *Bull. Mens. Inf. and liaison of the PNTTA* 68: 1- 4.
[4] Boujghagh M, and Chajia L, **2001**. *Earth and life* 52: 1-7 .
[5] Nobel, P. S, **1988,** in Barbera, G. ,Inglese,P. ,and Pimienta-Barrios , P. , (Eds.), Agro ecology, cultivation and uses of cactus pear (pp. 36- 45). FAO Plant Production and Protection, Paper: 132.
[6] Oppenheimer HR**, 1962***:* research on the arid zone water trade circles of arid and semi arid. Record of the research of UNESCO
[7] Sudzuki Hills, **1995**. Anatomy and morphology in Barbera G et al J (eds), agro-ecology, cultivation and uses of cactus pear, FAO. Plant product and protect Division Paper 132, Rome (Italy), 28-35.
[8] Kartez R. , **1996**. Nature. The book of Paris hachette printed in Italy by G. GANA.
[9] Mulas M., Mulas G., **2004**, potential of strategic use of plants of the genera atriplex and Opuntia in the fight against desertification, research group University of Sassari (Italy) http://desa.uniss.it/mulas/desert
[10] Nefzaoui A. , Ketata H. and Mohammed El Mourid M. ,**2012**, Agricultural Technological and InstitutionalInnovations for Enhanced Adaptation to Environmental Change in North Africa, *(ICARDA) North Africa Program, Tunis,Tunisia* www.intechopen.com
[11] Barbera G.**, 1995.** History,economic and agro-ecological importance of cactus. In:Barbera, G., Inglese, P., & Pimienta-Barrios , P. (Eds), agro-ecology, cultivation and uses of cactus pear. Eds FAO plant production and protection, p132.
[12] Sudzuki, F., Muñoz, C., Berger, H., **1993**. The culture of the prickly pear (Pear cactus). In Carmen Saenz (p 7 - 24), FAO Agricultural Services Bulletin N°162
[13] Nefzaoui, A. ,**1996**, nutritional value of spinless cactus (*Opuntia ficus indica)* and Atriplex (*Atriplex numularie*) based number for sheep, vol. II, proceedings of the workshop on native and exotic fodder shrubs in arids and semi-arids areas 27 Oct. - 2 Nov., 1996, Hammamet (Tunisia), (eds) INRAT/ICARDA/SWP/ Ministry of Agriculture **,** (**2001**), 518-523.
[14] Andrade-Montemayorhas, M., Cordova-Torresb A. V., Garcia-Gascac T., Kawasd J. R., **2011**, *Small Ruminant Research*, 98 (83- 92)
[15] Stintzing F. C., and R. Carle R, **2005**. *Mol. Nutr. Food Res.* ,49, 175 - 194
[16] Feugang. J. M, predicts. P, Zou. D, Stintzing. F. C and Changping Zou **2006**: *Frontiers in Bioscience* **; 11**, 2574-2589**,**

[17] Hadj Sadok T. , Ida F. , Bellal M. , Maria Stela Abdul Hussain, **2008**, *Agricultura, agricultural practice and science journal*, Vol 65, N°. 1-2
[18] Majid atmani, Fouad EL BOUAMI and Zoubida LAGHRARI Pour une étude de l'impact de la sécheresse sur le climat méditerranéen du massif de Moulay Driss Zerhoun (Prerif,Maroc). Geomaghrib, **2004** 2 pp119-125
[19] A. Cellar, Pharmacognosy, phytochemistry, medicinal plants. 2nd Ed. Tech. And Doc. Ed. Lavoisier, Paris, **1993**, 274-285.
[20] E. Trease, W. Evans,C. Pharmacognosy. Billiaire.Tindall.London 13 Edn,**1987**, 61-62.
[21] J. Memelink,R. Verpoort,J. W. Kigine,Organization of jasmonate responsive gene expression in alkaloid metabolism.2**0**01.
[22] G. Linden,D. Lovient,Biochemistry agro-industrial. Upgrading food agricultural production . Ed. Asson. Paris, **1994**, 104-109.
[23] Bruneton, J. Pharmacognosy, phytochemistry, medicinal plants. 3th Ed. Tec. e Doc. Eds. avoisier.Paris, **1999**, 199-388.
[24] J. B. Harborne, Phytochemical methods, London. Chapman and Hall, Ltd,**1973**, pp. 49-188.
[25] Guignard, J. L. Abstract of plant biochemistry. 2nd Ed. Masson.Paris, **1979** 84.
[26] AOAC, **1990**; Official Methods of analysis of association of official analytical chimists 13th Ed. AOAC. Washington., DC
[27] AOAC. **1984**.In: S. Williams, (Ed), 'Official Methods of Analysis' of the Association of Official Analytical Chemist International. 14th Ed. AOAC, Arlington, Virginia, USA,.
[28] Hayes, A. and B. Markovic, **2002**. *Food Chem. Toxicol*., 4: 949-964.
[29] Danova, S. , Petrov, K. , Pavlov, P. , Petrova, P. , **200**5. *Int. J. Dairy Technol* 58, 100-105. doi:10.1111 /j.1471-0307.**2005**.00194.x
[30] Diana-Roxana Pelinescu, Elena Sasarman, Mariana- Carmen Chifriuc , Ileana Stoica , Ana- Maria Nohit, Ionela Avram, Florea Serbancea, Tatiana Vassudimov, **2009**. *Rom. Biotechnol. Lett.*, Vol. 14, No. 2, 4225-4233
[31] Of Vuyst, Vanderveken, Van de Ven, Degeest, **1998**. *J. Appl. Microbiol*. 84, 1059-1068. doi:10.1046 /j.1365-2672.1998.00445.x
[32] Heim, Kelly E, Anthony R Tagliaferro, and Dennis J Bobilya. **2002**. *The Journal of Nutritional Biochemistry* 13 (10): 572-584
[33] Ghanim, H. , C. L. Sia, K. Korzeniewski, T. Lohano, S. Abuaysheh, A. Marumganti, A. Chaudhuri, and P. Dandona. **2011**. *endocrinology & metabolism* 96 (5) 1409-1414
[34] Tirilly and **Bourgeois1999** : Technology of vegetables ed. Tec and Doc Lavoisier p. 550;
[35] Hadj Sadok T, F. A. , **2014**. *Rev. Nat. Technol Natec B Sci. Agron. Biol. no*. 11, 17-19.
[36] FAO., (**1971**) "Techniques for developing pastoral", vol. 3, planting of fodder shrubs, FAO project Tun. 71/540, Ariana -Tunisia (1971), 17p.
[37] Combo, A.M. M. , Aguedo, M. , Paquot, M. , **2011**. *Biotechnol. Agron. About Society*. 15, 153-164.
[38] Sáenz, C., Sepúlveda, E., Matsuhiro, B., **2004**. *J. Arid approximately*. 57, 275-290. DOI:10.1016 /S0140-1963 (03)00106-X
[39] Pierre Luhata. SJ, E. Kalonda, A.B. Alonda, A.B. Kanangila, E. K. Kitwa, D. Mulungulungu, J. B. Lumbu Lumbu., **2008**. Chemical Study of the species Jacobinia Carnea . Memory Online. URL http://www.memoireonline.com/11/08/1619/ Etude-Chimique -de-lespece- Jacobinia-Carnea.html (accessed 6.26.15).
[40] Galati E,M, Monforte Mr T, Micelil N, Mondello MR,R Taviano Mr, F Galluzzo MR and Tripodo M,M, **2007**, *phytother Res* 21, 344-346
[41] Galati E. M. , ,. Monforte Mr. T. Tripodo, Mr. M, D'Aquino A. and Mondello MR. R. , **2001**. *J. of Ethnopharmacology* , 76, 1 , P 1-9
[42] Galati E. M. , Tripodo M. M. , Trovato A. , d'Aquino, A. , Monforte, M.T. , **2003** . *Pharmaceutical Biology*. 41, (3), 175-179
[43] Cardenas Medellin ML; Serna Saldivar S O? and Velazco of the Garza J, **1998** *Archivos latinoamercanos Nutricion :* 48, 316-323
[44] Lee, J. -C., Kim, H. -R. , Kim, J. , Jang, Y.-S. , **2002**. *J. Agric. Food Chem*. 50, 6490-6496.
[45] Leroux, J. , Langendorff, V., Schick, G., Vaishnav, V., Mazoyer, J., **2003**. Emulsion stabilizing properties of pectin. Food Hydrocoll, 6th International Hydrocolloids Conference 17, 455-462. DOI:10.1016 /S0268-005X(03)00027-4
[46] Abdessemed D. , Nezari M. , Mohamed Hadj A. R. , Abdessemed Dj, Bahadi F.and Halitim A., 2014. J. *Chem. Pharm.Res*. , **2014**, 6 (12) :198-201
[47] Jean Magloire Feugang, P.K.,**2006**. *Front. Biosci. J. Virtual Libr*. 11, 2574-89. doi:10.2741 /1992
[48] Sendra, E. , Fayos, P., Lario, Y., Fernandez-Lopez, J., Sayas-Barber á, E., Perez-Alvarez , J. A. , **2008**. *Food Microbiol* 25 , 13-21. doi:10.1016 /j.fm.2007.09.003