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Optimization of extraction in *Daphne gnidium* L. leaves

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ABSTRACT

Daphne gnidium L. is a medicinal plant used as hypoglycemic which is common in the western Mediterranean region. In our study we have focused on three methods of extractions which are used traditionally and we looked at the antioxidant potential responsible of its hypoglycemic activity and if it is related to the polar or non polar molecules. in all extracts and fractions we have measured Total phenols and Total saponosids. the capacity of scavenge of DDPH was measured in different fractions. Our results highlights the efficacy of fraction to exctract secondary metabolites and proof that the antioxidant activity of *Daphne gnidium* Leaves can come from polar and non polar molecules.

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INTRODUCTION

Nature has been a source of therapeutic agents for thousands of years, and an impressive number of modern drugs have been derived from natural sources, many based on their use in traditional medicine.The use of herbal drugs is once again becoming more popular in the form of food supplements, nutraceuticals, and complementary and alternative medicine

Daphne gnidium L. (*Thymeleaceae*), a common plant grows wild in Tessala Mounts (North-West Algeria) and the whole Algerian Tell, is confined mainly to the West Mediterranean, being relieved by *Daphne linearifolia* Hart in the Eastern part of the Basin (Quezel et santa,1963). This evergreen shrub with leathery leaves can grow to a height of 2 m (Zayat,1963).

The methanolic extract of *Daphne gnidium* L. leaves showed a presence of four coumarins (daphnetin, daphnin, acetyl- umbelliferon, and daphnoretin), nine flavonoids (apigenin, luteolin, quercetin, orientin, isoorientin, luteolin 7-*O*-glucoside, apigenin7-*O*-glucoside, genkwanin, and 5-*O*-β-D-primeverosylgenkwanine, and α-tocopherolwere) which contribute to its antioxidant activity(Deiana *et al*., 2003).Chaabane *et al.* (2012) have highlighted the quantification of phenolic compounds of leaf methanolic extract (total phenols = 157. 47 gallic acid equivalent, total flavonoids = 114.57 quercitin equivalents, tannins = 116 tannic acid equivalents).other study was effected by Dif *et al* to *Daphne gnidium* in the ecological and phytochemical context

The choice of extraction procedure depends on the nature of the source material and the compounds to be isolated. Prior to choosing a method, it is necessary to establish the target of the extraction (Satyajit *et al*.,2006)

Daphne gnidium is traditionally used as hypoglycemic due to the presence of polyphenols (zayate *et al*.,1997) but the traditional method of extraction is different between maceration, decoction and infusion, the present study looked in the difference between those methods and between the origine of antioxidant activity if it has an origin from polar molecules such as flavonoids or to non polar molecules such as triterpens

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MATERIAL AND METHODS

Biological material:

Daphne gnidium is collected from Tessala region (corodone, altitude) in august 2014 and then dried in the shield after that crushed and stored in closed boxes.

Extraction methods:

Firstly for maceration, we have used two different maceration by methanol and distilled water for this we have take 2 g of leaves powder then macerated in 20 ml of each solvent for 20 minutes than filtrated. secondly the decoction method is made by taking 2g of leaves powder in 20 ml of water then make it under 50°C for 20 minutes then filtrated. Thirdly, the infusion method is used by taking 2g of leaves in 20 ml of boiling water under ambient temperature for 20 minutes then filtrated

Fractions methods:

flavonoids extraction:

An alquilot of leaves powder are macerated in volume of methanol for 1 hour then evaporated and decanted in distilled water then washed by hexane in order to remove non polar content after that decanted with ethyl acetate (Harinder *et al*.,2006)

triterpen extraction:

An alquilot of leaves powder are decocted in distillated water for 1 hour then decanted with chloroform in order to remove polar fraction then decanted with ethyl acetate (Harinder *et al*.,2006)

phytochemical screening:

We have used NEU reagent (to determine the presence of flavonoids) and Lieberman-Burchard Reagent to determine the presence of triterpens) to each fraction of ethyl acetate (Albuquerque *et al*.,2014)

Antioxidant activity):

antioxidant activity of each fractions is measured using DPPH (diphenyl-1-picrylhydrazyl) method referring to Benhamou *et al*, 2006 the results is expressed by mg eq ascorbic acid/g of Dry matter . ascorbic acid used as a standard for the calibration curve (y=379,2x+5,25)

Total phenols assay:

A volume of 200 µl of the extract was mixed with 1 ml of Folin-Ciocalteu reagent diluted 10 times with water and 0.8 ml of a 7.5 % sodium carbonate solution in a test tube. After stirring and 30 min later, the absorbance was measured at 765 nm by using a Jenway 6405 UV-vis spectrophotometer. Gallic acid was used as a standard for the calibration curve. y=0.013X+0.(benhamou,2006)

Total saponosids assay:

We Place 250 µl of extract solution to 0.25 ml with 80% aqueous methanol. Then we add 0.25 ml of the vanillin reagent, and then 2.5 ml of 72% (v/v) sulfuric acid slowly on the inner side of the wall. after that we Mix the solution well and transfer the tubes to a water bath adjusted at 60°C. After 10 min, cool the tubes in ice-cold water for 3 to 4 min, and measure absorbance at 544 nm. Total saponosids compounds are reported as diosgenin equivalents by reference to a standard curve against the reagent blank (y= 0.00041X +0.1107) [11]

RESULTS AND DISCUSSION

Table 1: global results of *Daphne gnidium* leaves optimization

	Total phenols (mg GAE/gDM)	Total saponosids(mg DE/gDM)
decoction in water	6,64±0,047	4,87±1,03
infusion in water	6,24±0,33	5,73±0,17
maceration in water	5,72±0,39	5,48±1,21
maceration in methanol	15,36±0,19	58,17±1,21
ecetat extract	25,61±1,49	138,65±0,86

Our results (table 1) highlight that maceration using methanol has the best concentration of total phenols and total saponosids. In fact, the most important priority of extraction process is choosing the most appropriate solvent or system for the extraction of a particular target group involved in metabolism.

whereas maceration, infusion, decoction using water as solvent present approximate values and make about the half concentration of metabolites coming from maceration with methanol.

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Traditionally we have used infusion as suitable for the extraction of the weakest parts of the plant, in other hand decoction is widely used to extract the compounds of the Hard parts of plants (Albuquerque, 2014)

Generally, phenolic compounds are employed for the extraction of polar organic solvents (methanol, ethanol, or acetone) or a mixture of these solvents with water is utilised to optimise the process and to increase the yield whereas non polar molecules such as saponosids will infiltrated in that solvent (Harinder *et al*.,2006)

The fraction of tritrepens is commonly used from decoction extraction whereas the fraction of flavonoids is used from methanol maceration. In fact, the nature of the chemical species studied is unclear, one method to identify the phytochemical samples is the separation of the crude extract in solvents of increasing polarity; thus, the sample is semi purified in different partitions (Sarker, 2006). the primarly screening phytochimic (Albuquerque *et al*., 2014)

has validated the method of fraction to isolate flavonoids and triterpens. after that we have compare the antioxidant activity of each fraction, the results have shown approximately equal values: flavonoids (65,82± 0.9 mg EAA/g DM) tritrepens (65.72±0.9 mg EAA/g DM)

Conclusion:

Maceration with methanol is the best methods to extract total phenols and total saponosids this results validate the modern method of extraction of those molecules. Whereas, traditionally decoction method is the best method to extract total phenols at the temperature of 50 °C which is in the interval of the conservation of secondary metabolites

We hope to complete this work by the isolation and identification of polyphenols and triterpens and to test the hypoglycemic activity of each molecules using in vivo method.

REFERENCES

Albuquerque, U.P., V.F. Luiz, D.C. Cruz, F. Reinaldo, L. Paiva, R.N. Rômulo, 2014. Methods and Techniques in Ethnobiology and Ethnoecology.Humana press . edit springer pp: 151.

Benhammou, N., N. Atik Bekkara, T. Kadifkova Panovska, 2009. Antioxidant activity of methanolic extracts and some bioactive compounds of Atriplex halimus-C. *R. Chimie*, 12: 1259-1266.

Chaabane, F., J. Boubaker, A. Loussaif, A. Neffati, S. Kilani-Jaziri, K. Ghedira, L. Chekir-Ghedira, 2012. Antioxidant, genotoxic and antigenotoxic activities of *Daphne gnidium* leaf extract. *BMC Complementary and Alternative Medicine*, 12: 153-160.

Deiana, A., V. Rosa, F. Casu, L. Cottiglia, 2003. Bonsignore and M.A. Dessì, chemical composition and antioxidant activity of extracts from *Daphne gnidium* L *J. A.O.C.S.*, 80: 65-70.

Dif, M.M., F. Benali Toumi, M. Benyahia, A Benchohra Hadria A., 2014. Isolation of new flavonoid glycoside from *Daphne gnidium* L stems,. Adv. in Nat. Appl. Sci., 2(3): 4-6.

Dif, M.M., F. Benali Toumi, M. Benyahia, F. Koudache, 2014. First determination of physicochemical characteristic of *the Daphne gnidium* L. soil habitat in Tessala Mountain (NW of Algeria). Adv. in Nat. Appl. Sci., 2(3):1-3.

Dif, M.M., F. Benali Toumi, M. Benyahia, 2014. Effect of Passer Domesticus Ingestion on Daphne gnidium L. Seed Germination. Glob. J. Plant Ecophysiol, 4(1): 1-3.

Dif, M.M., F. Toumi Benali, M. Benyahia, K. Bouzid, M. Rahmani, F. Sellam, 2014. First determination of polyphenol variation in Daphne gnidium L. leaves depending on altitude and slope at Tessala Mounts (North-West Algeria). Revue écologie terre et vie. 69(4).

Harinder, P.S., P. Makkar, K.B. Siddhuraju, 2006. Plant Secondary Metabolites. Humana press . edit springer, pp: 142-144.

Quézel, P., S. Santa, 1963. *Nouvelle flore de l'Algérie et des régions désertiques méridionales*. C.N.R.S., Paris.

Satyajit, D., Z. Sarker, I. Lalexander, 2006. Natural Products Isolation. SECOND EDITION. Humana press pp: 121

Ziyyat, A., A. Legssyer, H. Mekhfi, A. Dassouli, M. Serhrouchni, W. Benjelloun, 1997. Phytotherapy of hypertension and diabetes in oriental Morocco. J. Ethnopharmacol, 58: 45-54.