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Isolation of new flavonoid glycoside from *Daphne gnidium* L stems

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ABSTRACT

Background: *Daphne gnidium* is plant that grows on Tessala mountain (western of Algeria). **Objective:** This plant is investigated by different research in the word and it is the first time to valorize this plant from this region.. **Results:** We have extract and identify Kaempferol 7-*O*-glucoside using NMR[1]H, [13]Cand UV methods**. Conclusion:** This molecule is firstly determind in this plant due to the novel method extraction used.

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INTRODUCTION

Numerous methods have been utilized to acquire compounds for drug discovery including isolation from plants and other natural sources, synthetic chemistry, combinatorial chemistry, and molecular modeling (Ley and Baxendale, 2002; Geysen *et al*., 2003; Lombardino and Lowe, 2004). Despite the recent interest in molecular modeling, combinatorial chemistry, and other synthetic chemistry techniques by pharmaceutical companies and funding organizations, natural products, and particularly medicinal plants, remain an important source of new drugs, new drug leads, and new chemical entities (NCEs) (Newman *et al*., 2000, 2003; Butler, 2004 ;Balunas et Kinghorn, 2005).

Flavonoids are a group of polyphenolic compounds diverse in chemical structure and characteristics. They occur naturally in fruit, vegetables, nuts, seeds, flowers, and bark and are an integral part of the human diet.

The aim of this work is flavonoids identification using RMN H1 C 13using co and UV method of *Daphne gnidium* L. stems (Thymeleaceae), a plant that commonly grows wild in Tessala (western Algeria) and we can found on the Mediterranean area and can grow to a height of 2 m (Ziyyat,1997). In folk medicine the infusion of the leaves is used as hypoglycemic (Bellakhdar, 1997) and to treat skin diseases (Bruneton,1997 ; Cardon,2003). This plant is also used in traditional textile dyeing (chaaban *et al*, 2012)

Methodology:

Extraction by different solvent:

initially We added 100 g of a fine powder of stems to volume mixture of ethanol / water (80:20) and then it'smacerated for three days and we renewed every 24 hours the solvent (350 ml x 3). Hydro-ethanolic solutions obtained are combined in a single container and then we have filtered to obtain a clear solution.

Then we evaporated the solution using a rotary evaporator (Rotavapor) R 120 at a temperature ranging between 35 with 45°C. We took again the dry extract by boiling water 200ml distilled, after we left during 24 hours in order to undergo a decantation, then it is filtered on filter paper Whatman n°1.

The clear aqueous phase after successive extractions is placed in a funnel to undergo successive confrontations with various solvents. We have used petroleum ether, chloroform, ethyl acetate and 1-butanol.for each stapes we have mixed 100ml of aqueous extract with each solvent than we have evaporated solution using a rotary evaporated then we had lyophilized the extract and we have found a brown sample

UV analysis:

Pure sample were measured with MeOH and with different diagnostic shift reagents (Mabry *et al*., 1970) on a UV IKON spectrophotometer.

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NMR analyses:

C13 and H1-NMR and analyses were run on Brucker 300, Jeol EX-270 and 400 MHz spectrometers relative to TMS in DMSO

Results:

Table 1: NMR data of new compound

	^{1}H	^{13}C
2	5.14 d	76.12
3	5.12 d	127.76
4		92.64
5	6.29	112.59
6	6.32	129.26
7	6.71 d	157.49
8	7.12 d	121.85
9	7.13	157.49
10		125.82
1’		150.70
2’	6.70	118.66
3’		145.25
4’	7.15 d	153.13
5’	6.70 d	134.53
6’		143.13
C=O		158.57
glucose		
1	3.70	74.70
2	3.14	72.04
3	3.62	73.49
4	3.83	62.57
5	3.14	70.25
6	3.78	61.61

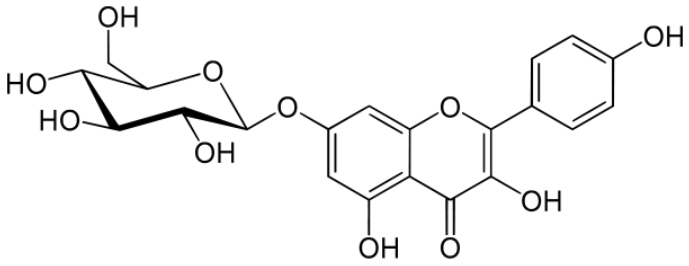

Fig. 1: chemical structure of Kaempferol 7-*O*-glucoside isolated from Daphne gnidium stems

Discussion:

Kaempferol-O-glucoside was isolated from the hydroethanolic extract of daphgne gnidium stem extract as a light brown powder. Its molecular formula was assigned to be C21H22O9 by the COSY and Tcocsy 2D NMR spectra.

The C 13spectrum, revealed thepresence of 21 carbons, consisting of one methylene, 9 methine, two methyl, and eight quaternary carbons. Unambiguous assignments were performed by heteronuclear shift correlation spectroscopy H1, C13 NMR and The UV–Vis absorption spectra of ,Indicated dihydroflavonol derivative with free hydroxyl groups at C-3 and C-5, and the observed shift in alkali suggested the 7-hydroxyl group to be substituted

UV (MeOH):kmax270-360 (Mabry, Markham, & Thomas, 1970). The H NMR spectrum showed two doublets at d5.14and 5.1(J= 12.1 Hz) characteristic of transH-2/H-3 protons in a dihydroflavonol. The proton at d6.29 correlated with the carbons atd97.6 and 112.29 in the T cocsy spectrum, therefore it was assigned to the H-6 and H-8 protons of ring A. From the T cocsy experiment, ring B was assigned as a 1,4-disubstituted benzene ring (7.16 d, 2H j =9.5Hzand 5.60 d, 2H j= 9.5Hz). The sugar functionality was identified as b-glucopyranose by the 1Hand13C spectral data.

Conclusion:

We had show a new method to extract Kaempferol-O-glucoside from Daphne gnidium stems

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Further investigations will be important to poof biological activity of this compound and his valorization on prevention and therapies

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