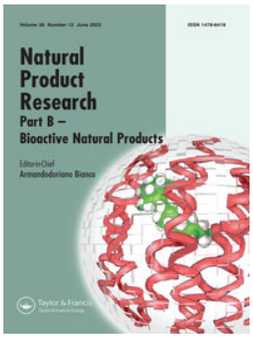

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SHORT COMMUNICATION

Chemical constituents, *in vitro* anti-inflammatory, antioxidant and hemostatic activities of the *n*-butanol extract of *Hyacinthoides lingulata* (Poir.) Rothm

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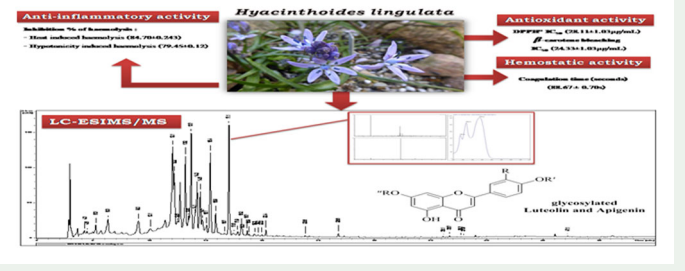
ABSTRACT

The phytochemical profile obtained from LC-ESI-MS/MS analysis of the *n*-butanol extract (BEHL) from the North African endemic plant *Hyacinthoides lingulata* (Poir.) Rothm. brought about the identification of ten glycosylated derivatives of apigenin and luteolin flavones. For the same plant extract, *in vitro* anti-inflammatory (hypotonic induced hemolysis and heat induced haemolysis assay) and antioxidant (DPPH and β-Carotene) activities were evaluated observing high inflammatory inhibition by protecting membrane stability of erythrocyte in both heat ($84.70 \pm 0.24\%$) and hypotonic induced hemolysis ($79.45 \pm 0.12\%$). A remarkable hemostatic effect was also established by measuring the coagulation time (15.95 ± 1.05 s at a dose of 1 mg/mL) of decalcified plasma related to its phytochemical content. It is the first report on combined chemical components and biological evaluation of this specific plant.



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1. Introduction

In Africa, the genus *Hyacinthoides* (Asparagaceae) is considered as a valuable source of medicinal plants used to treat women infertility and constipation (Breyer-Brandwijk and Watt 1962), and promoting blood circulation, as an anti-inflammatory agent and as an analgesic (Nishida et al. 2008). *Hyacinthoides lingulata* (Poir.) Rothm. (Syn: *Scilla lingulata* Poir.) is a small perennial plant originally endemic in Northern regions of Africa. In Algeria, it is called 'Becal el Far' and used for the treatment of menopause and gynecological problems (Chermat and Gharzouli 2015). Many biological studies revealed the glycosidase inhibitory (Watson et al. 1997), anti-inflammatory (Du Toit et al. 2011), anti-cancer (Ghoran et al. 2016) and antioxidant (Nishida et al. 2013) properties of some plants belonging to *Hyacinthoides* species. According to the literature, this genus is known for its containment of homoisoflavonoids, alkaloids, triterpenoids, stilbenoids, xanthone, lignin and phenylpropanoid glycoside (Mulholland et al. 2013; Fan et al. 2014).

The aim of the present study is to investigate the chemical composition and the biological properties of the *n*-butanol extract (BEHL) obtained from the whole endemic Algerian plant (bulbs, leaves, stems and flowers), for which no previous phytochemical and pharmacological data have been reported.

2. Results and discussion

LC-ESI-MS/MS analysis of BEHL led to the identification of the most abundant peaks detected in the corresponding UV chromatogram (Figure S1). According to data reported in Table S1, they were associated with luteolin 7-*O*-(rhamnosyl-(1→2)-glucoside)-3′ -*O*-(glucosyl-(1→2)-rhamnoside) (**1**), apigenin 7-*O*-(glucosyl-(1→2)-glucosyl-(1→6)-glucosyl-(1→2)-glucoside (**2**), luteolin 7-*O*-rhamnoside-3′-*O*-(glucosyl-(1→6)-glucoside) (**3**), apigenin 7-*O*-glucosyl-(1→2)-glucosyl-(1→6)-glucoside (**4**), apigenin 7-*O*-glucosyl-(1→2)-rhamnoside-(1→6)-glucoside (**5**), apigenin 7-*O*-glucosyl-(1→2)-glucosyl-(1→2)-rhamnoside(**6**), apigenin 7-*O*-(glucosyl-(1→2)-glucoside)-4′ -*O*-glucoside (**7**), luteolin 7-*O*-(glucosyl-(1→2)-glucoside)-3′ -*O*-rhamnoside (**8**), apigenin 7-*O*-(glucosyl-(1→2)-glucoside (**9**) and apigenin 7-*O*-(glucosyl-(1→2)-rhamnoside (**10**) (Figure S2). Compounds **1**–**8** and **10** were identified for the first time from the genus *Hyacinthoides*.

The total phenolic and flavonoid contents of BEHL were calculated according to the calibration curves established by gallic acid and quercetin, respectively. The results expressed in Table S2 show moderate phenolic and flavonoid contents with values of 97.56 ± 0.73 mg GAE/mg and 43.94 ± 0.51 mg QE/mg, respectively.

Under certain conditions the erythrocyte membrane can lead to the loss of its protective function and consequently cell lysis or hemolysis. BEHL inhibition on haemolysis of human red blood cell (HRBCs) induced by hypotonicity indicated a good result (Table S3) with a percentage of haemolysis inhibition (79.45 ± 0.12 at 400 μg/mL) close with the one of the anti-inflammatory drug indomethacin (84.82 ± 0.21 at 200 μg/mL). A similar behavior was observed with the doses able to protect the human erythrocyte membrane against lysis induced by heat (Table S4). These results provide evidence for

membrane stabilisation as an additional mechanism of anti-inflammatory activity, attributable to flavonoids, due to their ability to stabilise the membrane (Jorge 2004).

The exact mechanism of BEHL's effect on membrane stabilisation and the chemical constituents responsible for this effect so far are not known. However, a number of studies have shown that flavonoids (David 2007) and other plant compounds exhibit analgesic and anti-inflammatory effects due to their ability to stabilise the membrane (Mocan et al. 2014). The anti-inflammatory activity attributed to polyphenols can be carried out by several mechanisms, such as a decrease in the synthesis of pro-inflammatory mediators like IL-1β, IL-2, IL-6, IL-8, IFN-γ and TNFα, their release and decreased activity of immune cells (Teplova et al. 2018). In addition, polyphenols can inhibit the inflammatory response by targeting the inflammatory NF-κB pathway (Shen et al. 2017). On the other hand it has been shown that flavonoids have potential anti-inflammatory reagents because they inhibited proinflammatory cytokine-induced chemokine expression. It has been shown that apigenin and luteolin reduced the production of inflammatory mediators by inhibiting NF-κB activation (Funakoshi-Tago et al. 2011). Therefore, it is reasonable to postulate that the flavonoids in BEHL could be responsible for the membrane stabilising effect observed in our current study.

The antioxidant activity of BEHL was assessed using DPPH radical and β-carotene bleaching methods, indicating potent free radical-scavenging and antioxidant activities in comparison with Trolox and ascorbic acid taken as a standard (Table S5). The scavenging activity of DPPH increased proportionally with extract concentration and the IC_{50} value was recorded (28.11 ± 1.03 μg/mL) which was moderate in comparison with Trolox and ascorbic acid (3.85 ± 0.15; 7.03 ± 0.09 μg/mL, respectively). Moreover, the same tendency of inhibition was observed in the β-carotene bleaching assay with an IC_{50} value of 24.33 ± 1.03 μg/mL (trolox: 4.15 ± 0.23 μg/mL; ascorbic acid: 8.22 ± 0.11 μg/mL). All the results were statistically insignificant at 0.05 levels.

The coagulation which occurs in the mammalian plasma involves a large number of proteins causing the formation of thrombin; the enzyme then converts fibrinogen into an insoluble fibrin clot (Davie 1986). The evaluation of hemostatic activity of BEHL on the coagulation time have showed a dose-dependent effect plasma (Table S6), which means that the BEHL significantly delayed clotting time $p \leq 0.05$ more than the control. It is known that the hemostatic activity of plant extract is mainly due to the presence of polyphenols/flavonoids, which act as anticoagulants. According to the literature, coumarins are considered to be competitive inhibitors of vitamin K in the biosynthesis of prothrombin and prevent the conversion of the latter, which is a coagulation factor dependent on vitamin K to thrombin, and consequently prevention of thrombotic conditions represented by coagulation (Islam et al. 2017). Our results can be explained by the presence of flavonoids as natural source of anticoagulant agents. In particular, the investigation of different extracts from *Linaria reflexa* indicated the best coagulation time for *n*-butanol extract containing glycosyl flavonoids (Cheriet et al. 2019).

3. Conclusion

This study shows that *n*-butanol extract of *H. lingulata* (BEHL) has *in vitro* anti-inflammatory, antioxidant, and hemostatic activities. These activities may be majorly

attributed to its high presence of natural compounds, examined by LC-ESI-MS analysis. In light of this study findings, the effectiveness of *H. lingulata* as a medicinal plant is promising for further studies to discover its metabolites acting as pharmacological agents.

Disclosure statement

No potential conflict of interest was reported by the authors.

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References

Breyer-Brandwijk MG, Watt JM. 1962. The medicinal and poisonous plants of Southern and Eastern Africa. 2nd ed. Edinburgh: E. and S. Livingstone Ltd.

Cheriet T, Hanfer M, Mancini I, Benelhadj S, Laouas NE, Ameddah S, Menad A, Seghiri R. 2019. Anti-inflammatory and hemostatic effects of *Linaria reflexa* Desf. Nat Prod Res. :1–6. https://doi.org/10.1080/14786419.2019.1663516

Chermat S, Gharzouli R. 2015. Ethnobotanical study of medicinal flora in the North East of Algeria - An empirical knowledge in djebelzdimm (Setif). J Mater Sci Eng A. 5:50–59.

David S. 2007. Studies force new view on biology of flavonoids. Biol Med. 541:737–787.

Davie EV. 1986. Introduction to the blood coagulation cascade and cloning of blood coagulation factors. J Protein Chem. 5(4):247–253.

Du Toit K, Kweyama A, Bodenstein J. 2011. Anti-inflammatory and antimicrobial profiles of *Scilla nervosa* (Burch.) Jessop (Hyacinthaceae. S Afr J Sci. 107(5/6):96–100.).

Fan MY, Wang YM, Wang ZM, Gao HM. 2014. Advances on chemical constituents and pharmacological activity of genus *Scilla*. China J Chin Mater Med. 39:162–170.

Funakoshi-Tago M, Nakamura K, Tago K, Mashino T, Kasahara T. 2011. Anti-inflammatory activity of structurally related flavonoids, apigenin, luteolin and fisetin. Int. Immunopharmacol. 11: 1150–1159.

Ghoran SH, Saeidnia S, Babaei E, Kiuchi F, Hussain H. 2016. *Scilla persicene*: a new homoisoflavonoid with cytotoxic activity from the bulbs of *Scilla persica* HAUSSKN. Nat Prod Res. 30: 1309–1314.

Islam T, Yu X, Badwal TS, Xu B. 2017. Comparative studies on phenolic profiles, antioxidant capacities and carotenoid contents of red goji berry (*Lycium barbarum*) and Black goji berry (*Lycium ruthenicum*). Chem Cent J. 11:59.

Jorge RM, Leite JP, Oliveira AB, Tagliati CA. 2004. Evaluation of antinociceptive, anti-inflammatory and antiulcerogenic activities of *Maytenu silicifolia*. J Ethnopharmacol. 94:93–100.

Mocan A, Vlase L, Vodnar D, Bischin C, Hanganu D, Gheldiu A-M, Oprean R, Silaghi-Dumitrescu R, Crișan G. 2014. Polyphenolic content, antioxidant and antimicrobial activities of *Lycium barbarum* L and *Lycium chinense* Mill leaves. Molecules. 19(7):10056–10073.

Mulholland DA, Schwikkard SL, Crouch NR. 2013. The chemistry and biological activity of the Hyacinthaceae. Nat Prod Rep. 30:1165–1210.

Nishida Y, Eto M, Miyashita H, Ikeda T, Yamaguchi K, Yoshimitsu H, Nohara T, Ono M. 2008. A new homostilbene and two new homoisoflavones from the bulbs of *Scilla scilloides*. Chem Pharm Bull. 56:1022–1025.

Nishida Y, Wada K, Toyohisa D, Tanaka T, Ono M, Yasuda S. 2013. Homoisoflavones as the antioxidants responsible from bulbs of *Scilla scilloides*. Nat Prod Res. 27:2360–2362.

Shen T, Han XZ, Wang XN, Fan PH, Ren DM, Lou HX. 2017. Protective effects of dietary polyphenols in human diseases and mechanisms of action. In: Al-Gubory K, Laher I, editors. Nutritional antioxidant therapies: Treatments and perspectives. Cham: Springer (Suisse); p. 321–323.

Teplova VV, Isakova EP, Klein OI, Dergachova DI, Gessler NN, Deryabina YI. 2018. Natural polyphenols: Biological activity, pharmacological potential, means of metabolic engineering (Review). Appl Biochem Microbiol. 54(3):221–237.

Watson AA, Nash RJ, Wormald MR, Harvey DJ, Dealler S, Lees E, Asano N, Kizu H, Kato A, Griffiths RC, et al. 1997. Glycosidase-inhibiting pyrrolidine alkaloids from *Hyacinthoides non-scripta*. Phytochemistry. 46(2):255–259.