行政院國家科學委員會專題研究計畫成果報告

Isoflavanquinone 及 Gingerdione 衍生物之合成及其抗血小板、抗炎與抗過敏活性(III)
Synthesis and Antiplatelet, Anti-inflammatory and Antiallergic Activities of Isoflavanquinone and Gingerdione Derivatives (III)

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一、中文摘要

為了得到強力的抗血小板、抗發炎與抗過敏之新型先導化合物，所以著手進行 substituted isoflavanquinone ($VI_{1\sim7}$) 與其相關化合物之合成及生物活性之評估。

主要的中間體 substituted 2'-benzyloxyisoflavone ($IV_{1\sim7}$) 係由 substituted 2-benzyloxy-2'-hydroxychalcone ($II_{1\sim7}$) 以 thallium (Ⅲ) nitrate 及 trimethyl orthoformate 處理，經由氧化性重排而獲得。繼而以 Pd/C 之催化性氫化反應將化合物 $IV_{1\sim7}$ 還原成相對應的 substituted 2'-hydroxyisoflavan ($V_{1\sim7}$)。最後利用 Fremy's salt 將 $V_{1\sim7}$ 氧化成 substituted isoflavanquinone ($VI_{1\sim7}$)。另一方面，將化合物$IV_{1\sim7}$以HBr debenzylation，再利用Fremy's salt進行氧化即可得到 substituted isoflavonequinones ($VIII_{1\sim7}$)。

將目標化合物 substituted isoflavanquinone ($VI_{1\sim7}$) 及其相關化合物進行抗血小板、抗發炎與抗過敏之活性評估，這些化合物中以具有 quinonyl 官能基之衍生物 substituted isoflavanquinone ($VI_{1\sim7}$) 及 substituted isoflavonequinones ($VIII_{1\sim7}$) 為較具潛力之物質，可做為新型抗血小板、抗發炎及抗過敏藥物之基本架構。

由薑(*Zingiber officinale*)分離出 gingerol、gingerdione 及 isodehydrogingerdione 之化學成分[17]，發現這些成分有很強之抗血小板及抗發炎活性。

合成二系列 cinnamic acid 及 gingerdione 類衍生物以發現新型抗血小板藥物。經抗血小板活性篩選發現，其主要抑制花生四烯酸所誘導之血小板凝集，同時發現這類化合物初步的結構與活性關係。其中 [5]-gingerdione 活性最為顯著，甚至超越 indomethacin。但是其對於 cyclooxygenase 的抑制作用卻與 indomethacin 大不相同。

關鍵字：異黃烷苯醌、異黃酮苯醌、薑酮、抗血小板活性、抗發炎活性、抗過敏活性

Abstract

In order to obtain novel lead compounds with potent antiplatelet, anti-inflammatory and antiallergic activities, substituted isoflavanquinones and its related compounds were synthesized and evaluated for bioactivities.

Substituted 2'-benzyloxyisoflavone ($IV_{1\sim7}$), the key intermediate, was synthesized through the oxidative rearrangement of substituted 2-benzyloxy-2'-hydroxychalcone ($II_{1\sim7}$) with thallium (Ⅲ) nitrate. The products $IV_{1\sim7}$ were then reduced by catalytic hydrogenation on Pd/C to the corresponding

substituted 2'-hydroxyisoflavan ($V_{1\sim7}$) with loss of the benzyl group. Compounds $V_{1\sim7}$ were further oxidized with Fremy's salt to give the target compounds substituted isoflavanquinone ($VI_{1\sim7}$). Compounds $IV_{1\sim7}$ were debenzylated by HBr and then oxidized with Fremy's salt to give substituted isoflavonequinones ($VIII_{1\sim7}$).

The target compounds $VI_{1\sim7}$ and their related compounds were evaluated for their antiplatelet, anti-inflammatory and anti-allergic activities. Among compounds tested, 3-quinonyl derivatives, namely substituted isoflavanquinone ($VI_{1\sim7}$) and substituted isoflavonequinones ($VIII_{1\sim7}$) were the better promising agents, which provide a novel structural prototype for antiplatelet, anti-inflammatory and antiallergic agents.

Gingerol, gingerdiones and isodehydrogingerdione were isolated from *Zingiber officinale*. Some of these natural products have been found to process potent antiplatelet and anti-inflammatory actives.

Two series of cinnamic acid derivatives and gingerdione analogs were synthesized in search of novel antiplatelet agents. Screening for antiplatelet activity confirmed their inhibitory effects preliminary against AA-induced platelet aggregation. Among them, [5]-gingerdione showed the greatest potency that was even superior to indomethacin. However, its inhibitory effects on cyclo-oxygenase was very different from indomethacin.

Keywords : Isoflavanquinone, Isoflavonequinone, Gingerdione, Antiplatelet activity, Anti-inflammatory, Antiallergic activity

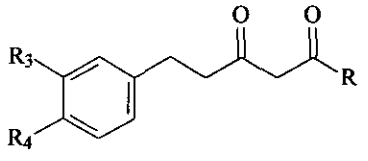
二、緣由與目的

經過去兩年的研究，本研究室合成了 isoflavanquinone， isoflavonequinone 及 3'-methoxyflavanquinone，經藥理活性測試之後發現此三種具有 quinone B 環之化合物對抗血小板凝集、中性白血球脫顆粒反應、中性白血球過氧化物形成、肥胖細胞脫顆粒反應等同時具有強力之抑制活性，是值得深入探討之領域。

於是繼續合成 substituted isoflavanquinones 衍生物 ($VI_{1\sim7}$)，並利用這些化合物及其相關化合物之生理活性數據，加以比較歸納以尋求生理活性更優越之化合物，提供進一步藥理作用之評估，亦可做為新型先導化合物 (Lead compound) 以供今後進一步化學結構修飾之用。

Gingerol類成分對AA所引起之血小板凝集具有相當強之抑制活性，然而其結構類似之gingerdione雖有對PG bio-synthetase抑制活性之報告，但是其對血小板凝集之影響卻未見諸於文獻。

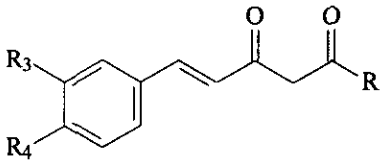
在本研究中著者仿照先前 ferulamide 類似化合物之研究模式以[8]-gingerdione (**22**) 及[10]-gingerdione (**24**)為先導化合物[13,14]，在通式 I 結構上 R，R_3，R_4 三個位置做適當的修飾，合成一系列之 1-(4-hydroxy-3-meyhoxyphenyl)-3, 5-alkanedione 類衍生物，另一方面亦合成了一系列通式 II之 2, 3-unsaturated 衍生物將兩大類 gingerdione 類衍生物提供抗血小板活性之篩選，建立其結構與活性關係進而選擇對 AA 所引起之血小板凝集抑制活性強之化合物測試其對之 COX-1，COX-2 抑制活性[18,19,20]，以期尋求作用機轉特異之新型抗血小板活性物質。



[8]-gingerdione R = $(CH_2)_6CH_3$, R_3 = OCH_3, R_4 = OH

[10]-gingerdione R = $(CH_2)_8CH_3$, R_3 = OCH_3, R_4 = OH

I R = $(CH_2)_nCH_3$, n = 0-8
R_3 = H, OCH_3
R_4 = H, OCH_3



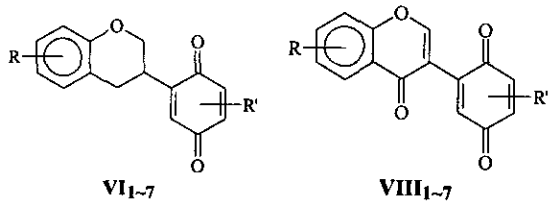
R = $(CH_2)_nCH_3$, n = 0-8
II R_3 = H, OCH_3
R_4 = H, OCH_3

三、結果與討論

I. Isoflavanquinones 及相關化合物之合成及其生理活性:

I-1. Substituted Isoflavanquinones ($VI_{1\sim7}$) 及Substituted Isoflavonequinones ($VIII_{1\sim7}$) 之合成

以 substituted 2-hydroxybenzaldehyde 為起始原料，經 6 個步驟即可得到標的化合物 ($VI_{1\sim7}$)，$VI_{1\sim7}$ 分別是 7-methoxy-isoflavanquinone (VI_1)，6-fluoroisoflavan-quinone (VI_2)，4'-methoxyisoflavanquinone (VI_3)，6-methylisoflavanquinone (VI_4)，6-chloroisoflavanquinone (VI_5)，5-methoxy-isoflavanquinone (VI_6)，6,8-dichloroiso-flavanquinone (VI_7)。另一方面，將化合物 $IV_{1\sim7}$ 以 HBr debenzylation，再利用 Fremy's salt 進行氧化即可得到 Substituted Iso-flavonequinones ($VIII_{1\sim7}$)，$VIII_{1\sim7}$ 分別是 7-methoxyisoflavonequinone ($VIII_1$)，6-fluoroisoflavonequinone ($VIII_2$)，4'-me-thoxyisoflavonequinone ($VIII_3$)，6-methyl isoflavonequinone ($VIII_4$)，6-chloro-iso-flavonequinone ($VIII_5$)，5-methoxyiso-flavonequinone ($VIII_6$)，6,8-dichloroiso-flavonequinone ($VIII_7$)。



$VI_{1\sim7}$ $VIII_{1\sim7}$

I-2. Substituted Isoflavanquinones ($VI_{1\sim7}$) 及 Substituted Isoflavonequinones ($VIII_{1\sim7}$)及其中間體之生理活性

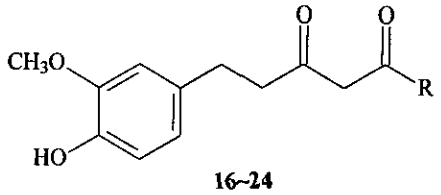
已經完成藥理活性測試的化合物中，以 7-methoxyisoflavanquinone (VI_1)，6-fluoro-isoflavanquinone (VI_2)，4'-methoxy-isoflavanquinone (VI_3)，7-methoxyiso-flavonequinone ($VIII_1$)，6-fluoroisoflavone-quinone ($VIII_2$) 及 4'-methoxyisoflavone-quinone ($VIII_3$)等具有 quinone B 環之化合物對抗血小板凝集 [3,4]、中性白血球脫顆粒反應 [5,6,7,8]、中性白血球過氧化物形成 [6]、肥胖細胞脫顆粒反應 [5,9,10,11,12] 等同時具有強力之抑制活性。其餘尚未討論之化合物，則仍在測試藥理活性中。

II. Cinnamic acid derivatives and gingerdione analogs 之合成及藥理結果

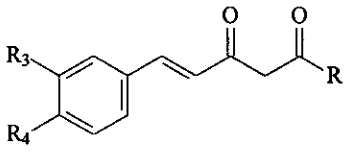
II-1. 合成

以 Ferulic acid (1)為起始原料 [15,16]，先經 catalyst hydrogenation，然後進行酯化，再將 phenolic OH 保護形成化合物，加水分解成酸，繼而用 $SOCl_2$ 處理使形成 ferulyl chloride (5)，然後在 LDA 存在下與各種 ketone 反應形成 diketone (7~15)衍生物，最後以還原方法將保護基 benyl group 去除，即可得到標的化合物 (16~24)。

以 cinnamic acid analogs (25~27)為起始原料，直接用 $SOCl_2$ 處理使形成 ferulyl chloride (28~30)，或經 catalyst hydrogenation 後(37~39)，再用 $SOCl_2$ 處理使形成 chloride (40~42)，然後在 LDA 存在下與各種 ketone 反應形成 diketone 衍生物 (31~36, 43~48)。

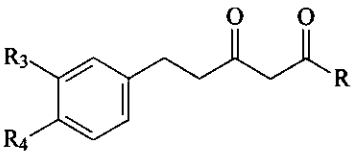


16~24
16 R = CH_3
17 R = CH_2CH_3
18 R = $(CH_2)_2CH_3$
19 R = $(CH_2)_3CH_3$
20 R = $(CH_2)_4CH_3$
21 R = $(CH_2)_5CH_3$
22 R = $(CH_2)_6CH_3$
23 R = $(CH_2)_7CH_3$
24 R = $(CH_2)_8CH_3$



31~36

31 R_3=R_4= OCH_3, OCH_3, R = $(CH_2)_6CH_3$
32 R_3=R_4= OCH_3, OCH_3, R = $(CH_2)_8CH_3$
33 R_3= OCH_3, R_4=H, R = $(CH_2)_6C$
34 R_3= OCH_3, R_4=H, R = $(CH_2)_8CH_3$
35 R3=R4=H, R = $(CH_2)_6CH_3$
36 R3=R4=H, R = $(CH_2)_8CH_3$



43~48

43 R_3=R_4= OCH_3, OCH_3, R = $(CH_2)_6CH_3$
44 R_3=R_4= OCH_3, OCH_3, R = $(CH_2)_8CH_3$
45 R_3= OCH_3, R_4=H, R = $(CH_2)_6C$
46 R_3= OCH_3, R_4=H, R = $(CH_2)_8CH_3$
47 R3=R4=H, R = $(CH_2)_6CH_3$
48 R3=R4=H, R = $(CH_2)_8CH_3$

II-2.藥理結果

抗血小板活性：

所合成之 gingerdione 及 cinnamic acid 衍生物部份生理活性試驗結果發現結構上 phenyl 之第 3 位為 methoxyl group 及第 4 位為 hydroxyl group 時，對所引起之血小板凝集抑制作用較為明顯。又當 n 逐漸增大，由 methyl group (**16**) 逐漸增大到 butyl group (**19**)時，其活性也隨著 alkyl group 之增大而增強。但是當 alkyl group 增大到 $(CH_2)_4CH_3$ 以上時 (**20-24**)，其強度就呈現下降之現象。

至於 gingerdione 衍生物對所引起之血小板凝集之抑制活性以化合物 **19** 之活性最強，約相當於 indomethacin 之 10 倍，是相當值得進一步探討之活性物質。於是將化合物 **16~24** 測試其對 COX-1，COX-2 之抑制活性，結果發現這類化合物對 COX-1，COX-2 均呈現相當弱之活性，其 IC_{50} 大於 300μM，此現象與 indomethacin 或 aspirin 顯然不同，其作用機轉值得進一步的探討。

四、計畫成果自評

本研究的內容與原計畫相符，亦幾乎完成預定之工作項目，研究之成果具學術價值，將在學術期刊發表或申請專利，雖然 quinone 類標的化合物之活性強度都比未取代之 isoflavanquinone 及 isoflavone-quinone 為弱，但都比 positive control 之活性更強，其藥理活性亦值得再做進一步地探討。

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